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Biotic and abiotic factors contributing to the invasion success of microstegium vimineum in eastern deciduous forests.

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BIOTIC AND ABIOTIC FACTORS CONTRIBUTING TO THE INVASION SUCCESS OF *MICROSTEGIUM VIMINEUM* IN EASTERN DECIDUOUS FORESTS

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M.S.S.W. University of Louisville 2010

A Dissertation

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ABSTRACT

BIOTIC AND ABIOTIC FACTORS CONTRIBUTING TO THE INVASION SUCCESS OF *MICROSTEGIUM VIMINEUM* IN EASTERN DECIDUOUS FORESTS

Kimberly Koenig

December 2, 2022

Forest managers face multiple challenges in maintaining woodland systems, including climate change, habitat destruction and fragmentation, and the invasion of novel species. Invaders can change microclimates, alter nutrient cycling and understory habitat, and outcompete native species, leading to native species population declines and reduction in species richness. To effectively combat plant invasions it is necessary to first understand factors contributing to invasive species spread, including the complex interactions between invaders and native biota.

This dissertation investigates the abiotic conditions and biotic interactions associated with invasion success of the non-native grass *Microstegium vimineum*, which is a species of great concern for forest managers throughout the eastern US. My initial study identified the most important abiotic elements associated with *Microstegium* presence. I found light availability and soil moisture to be the strongest predictors of *Microstegium* cover, followed by soil nitrogen and soil phosphorous. In this study I also examined the relationship between *Microstegium* and soil arbuscular mycorrhizae. I found no differences in abundance of soil mycorrhizae between invaded and noninvaded areas, and no difference in root colonization of *Microstegium* across abiotic gradients.

My second study analyzed the germination and growth of *Microstegium*, native grasses, and native woody species in field soils associated with *Microstegium* invasion. I found that *Microstegium*-associated soils enhanced *Microstegium* seed germination, while inhibiting the germination of native grass species. Surprisingly, I found greater aboveground biomass and stem height of native woody species in *Microstegium*-associated soils; however, I also found increased seedling mortality in *Microstegium*-associated soils.

For my final study, I surveyed *Microstegium* invasion at the landscape scale in seven local nature reserves to create a habitat suitability analysis using Geographic Information System (GIS) processing. I combined various habitat variables indicative of *Microstegium* presence to identify areas of greatest habitat suitability. These findings can inform best practices for prevention and eradication of *Microstegium*. For example, the creation of a habitat suitability analysis for a nature reserve, coupled with the knowledge of the importance of various abiotic factors to *Microstegium* presence, can assist land managers to direct limited resources to areas of highest susceptibility to invasion or to mitigate population spread.

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CHAPTER I

INTRODUCTION

One of the greatest threats to Eastern deciduous forests is invasion by plants facilitated by human activity (Kerns et al. 2020). Invaders can change microclimates (Watling et al. 2011, Ruckli et al. 2013, Setterfield et al. 2018), alter nutrient cycling (Levine et al. 2003, Liebhold et al. 2017), alter understory habitat (Medley 1997, Hartman and McCarthy 2008), and outcompete native species (Baiser et al. 2008, Johnson et al. 2015), leading to native species population declines (Flory and Clay 2010b, Aronson and Handel 2011, Flory et al. 2017) and reduction in species richness (Oswalt et al. 2007, Adams and Engelhardt 2009, Flory and Clay 2009, Droste et al. 2010, Flory and Clay 2010a, Beasley and McCarthy 2011). Loss of biodiversity in deciduous forests results in irrevocable changes to ecosystem functioning (Thompson and Starzomski 2007, Wardle et al. 2011), furthering the loss of native habitat already threatened by climate change, fragmentation, and removal of woodland areas for commercial, housing, and agricultural establishments. While ongoing research continues to provide insights into invasive species success, the complexity of associations between plant invaders and native communities creates a tangled web of interactive consequences, confounding our understanding of how best to protect our natural areas.

Over twenty hypotheses have been proposed to explain invader success (Enders et al. 2018), some conflicting and others synergistic (Jeschke 2014, Lau and Schultheis

2015), and many dependent upon the system and the plant invader (Jeschke et al. 2012). From these, several factors have emerged as probable contributors to plant invasion success. Novel associations with soil organisms such as arbuscular mycorrhizal fungi can provide an invader with advantageous access to resources (Reinhart and Callaway 2006), or the lack of association can lead to a reduction of mycorrhizal abundance and diversity, inhibiting native plant species (Vogelsang and Bever 2009). Invaders can change soil chemistry through altered nutrient cycling (Ehrenfeld et al. 2001, McGrath and Binkley 2009, Lee et al. 2012), by increasing uptake of specific nitrogen forms (Ehrenfeld et al. 2001, Fraterrigo et al. 2011, Ross et al. 2011, Lee et al. 2012), altering soil pH through exudates (Kourtev et al. 1998, Ehrenfeld et al. 2001, McGrath and Binkley 2009, Strickland et al. 2011, Lee et al. 2012), and trapping carbon and nitrogen resources in biomass litter with slower decomposition rates (Ehrenfeld et al. 2001, McGrath and Binkley 2009, Strickland et al. 2011, Kramer et al. 2012). Invaders also outcompete native species via early spring emergence (Smith 2013) and ruderal growth patterns (Martin et al. 2009) allowing better access to light (Baiser et al. 2008), and through copious seed production (Gibson et al. 2002, Warren et al. 2012), allowing for swifter population growth and dispersal ability (Barden 1987, Gibson et al. 2002).

Microstegium vimineum is an annual grass native to Asia and introduced to North America in the early 1900's (Fairbrothers and Gray 1972). This species demonstrates several novel traits: as a C4 species it thrives in open sun conditions (Touchette and Romanello 2010, Ziska et al. 2015); however, it can also survive and reproduce in shaded areas typically occupied by C3 grasses (Winter et al. 1982, Claridge and Franklin 2002, Cheplick 2005). In Kentucky, *Microstegium* is the only recorded C4 grass species found within forest interiors (Campbell, 2004). Developmental plasticity (Gibson et al. 2002, Droste et al. 2010, Cheplick 2015) combined with chasmogamous and cleistogamous seed production (Gibson et al. 2002, Cheplick 2005, Huebner 2011, Ziska et al. 2015) contribute to the success of *Microstegium* in eastern deciduous forests.

At the landscape level, *Microstegium* populations are known to spread along disturbance routes such as roads, trails, and streams (Christen and Matlack 2009, Miller and Matlack 2010, Ziska et al. 2015, Rauschert et al. 2017), establishing pioneer populations in open edge habitats (Barden 1987, Cheplick 2010, Warren et al. 2011b) which can then migrate into the forest interior (Huebner 2010). At the community level, *Microstegium* can outcompete native tree species, resulting in diminished forest regeneration (Baiser et al. 2008, Flory and Clay 2010a,b, Aronson and Handel 2011). *Microstegium* is known to alter nitrogen cycling (Fraterrigo et al. 2011, Ross et al. 2011) through preferential uptake of nitrate (Ehrenfeld et al. 2001, Lee et al. 2012), alter soil pH to a more basic level (Kourtev et al. 1998, McGrath and Binkley 2009, Strickland et al. 2011, Lee et al. 2012), and sequester carbon in annual litter (Strickland et al. 2011, Kramer et al. 2012). *Microstegium* invasion also increases the intensity of seasonal fires, causing the fires to burn at higher temperatures and for longer periods of time, suppressing native tree seedling survival (Flory et al. 2015).

Microstegium is also associated with changes to the abundance, function, and diversity of soil microbial organisms (Kourtev et al. 2002, Fraterrigo et al. 2011, Kramer et al. 2012, Shannon et al. 2012, North and Torzilli 2017), which may alter germination and growth of co-occuring native species. However, while *Microstegium* is known to associate with arbuscular mycorrhizal fungi (Lee et al. 2014), no studies to date have documented changes to this key group of soil organisms associated with *Microstegium* invasion.

STUDY SYSTEM

This dissertation research was conducted at ten forest reserve sites near Louisville, Kentucky, USA (Table 1, Fig. 1). The sites contained oak-hickory dominated forest systems, with common native species including *Acer* spp. (maple), *Quercus* spp. (oak), *Carya* spp. (hickory), *Fraxinus* spp. (ash), *Diospyros* spp. (persimmon), *Asimina triloba* (pawpaw), *Parthenocissus quinquefolia* (Virginia creeper), *Impatiens capensis* (jewelweed), *Ageratina altissima* (white snakeroot), and *Muhlenbergia schreberi* (nimblewill). Sites varied in size (66-6530 ha) and underlying geology (sandstone or limestone). All sites are located within the Interior Plateau Level III ecoregion and four different Level IV ecoregions (Table 1).

ORGANIZATION OF DISSERTATION

I explored the abiotic and biotic conditions which contribute to the invasion success of *Microstegium vimineum*. I used a combination of field studies, laboratory investigation, and GIS analysis.

In chapter two of this dissertation, I identified the most important abiotic and biotic factors contributing to *Microstegium* plant size and abundance in six local forest sites. I also characterized changes to the abundance of arbuscular mycorrhizal fungal (AMF) hyphae in the soil and to AMF root colonization of *Microstegium* along an abiotic gradient. Using a model averaging approach, I found that canopy openness (high light availability) and increased soil moisture were the strongest predictors of *Microstegium* cover, followed by soil phosphorous and soil nitrogen. Soil nitrogen was the sole significant predictor of *Microstegium* tiller height, although soil AMF abundance and percent root colonization of *Microstegium* by AMF were also included in the best fit model. Soil pH was excluded as a predictive factor in both analyses. In this study I also found no differences in the abundance of soil AMF hyphae between areas invaded by *Microstegium* and areas inhabited by native flora. While this study confirmed an association between *Microstegium* and arbuscular mycorrhizal fungi, I found no variation in root colonization density or extraradical hyphal growth across abiotic gradients.

In chapter three of this dissertation, I investigated the influence of soil associated with dense stands of *Microstegium* versus native vegetation on the germination and growth of *Microstegium*, four native grass species, and seven native woody species. I found *Microstegium* germination to be enhanced in *Microstegium*-associated soils, while germination of native grasses was inhibited. Woody species exhibited no difference in germination between soil types, but showed higher seedling mortality in *Microstegium*-conditioned soils. Unexpectedly, I found increased aboveground biomass and stem height for at least two of native species in soils associated with *Microstegium*. These results demonstrate contrasting responses to *Microstegium* invasion depending on plant habit.

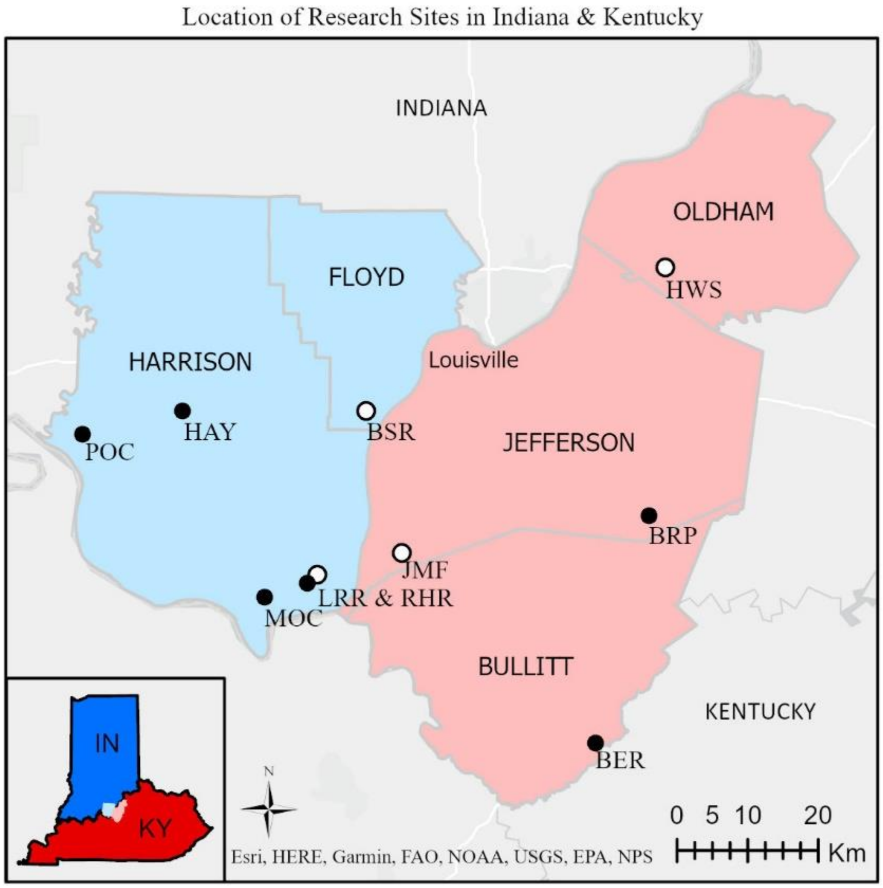
In chapter four, I used Geographic Information System (GIS) suitability analysis to classify areas of highest to lowest habitat suitability for *Microstegium*. I mapped populations and observed percent cover of *Microstegium* across seven field sites near Louisville, KY. I then obtained and modified online landscape data for GIS analysis: elevation, canopy cover, and distance from streams, roads, and trails. Additionally, I was able to derive a Tropical Wetness Index (TWI) layer as a proxy for soil moisture levels. Using known *Microstegium* habitat preferences, each data layer was reclassified from most to least suitable and added together to create an output map of suitable habitat. When combined with field data showing current locations of *Microstegium* populations, this tool will allow managers to best direct limited resources towards mitigation of dispersal in the areas of greatest risk for population expansion or colonization.

The fifth and final chapter of this dissertation summarizes the main conclusions and discusses ongoing and future research directions, such as using drone imagery to map *Microstegium* populations and examining the role of seed predation in preventing tree regeneration in areas heavily invaded by *Microstegium*.

Table 1: List of research sites near Louisville, KY.

Property Name	Owner/Manager	Location	Size of Property	Level IV Ecoregion
Bernheim Arboretum and Research Forest	Bernheim Arboretum	Bullitt County, KY	6530 ha	71c Knobs-Norman Uplands
Broad Run Park	The Parklands of Floyds Fork	Jefferson County, KY	242 ha	71d Outer Bluegrass
Brock-Sampson Nature Preserve	Indiana DNR	Floyd County, IN	247 ha	71c Knobs-Norman Uplands
Hayswood Nature Reserve	Indiana DNR & Harrison County Parks	Harrison County, IN	125 ha	71b Mitchell Plain
Horner Bird & Wildlife Sanctuary	University of Louisville	Oldham County, KY	85 ha	71d Outer Bluegrass
Jefferson Memorial Forest	Louisville Metro Parks	Jefferson County, KY	2630 ha	71c Knobs-Norman Uplands
LRR: Private Property	Property Owner	Harrison County, IN	3.6 ha	71b Mitchell Plain
Mosquito Creek Glades	The Nature Conservancy	Harrison County, IN	414 ha	71b Mitchell Plain
Post Oak Cedar Reserve / Harrison-Crawford State Forest	Indiana DNR	Harrison County, IN	91 ha	71a Crawford Uplands
Rabbit Hash Ridge Nature Reserve	The Nature Conservancy	Harrison County, IN	66 ha	71b Mitchell Plain

Figure 1: Location of research sites near Louisville, KY. White dots indicate sites used in multiple chapters, while black dots indicate sites used in only one chapter. Site codes are as follows: BER: Bernheim Arboretum and Research Forest, BRP: Broad Run Park at the Parklands of Floyds Fork, BSR: Brock-Sampson Nature Preserve, HAY: Hayswood Nature Reserve, HWS: Horner Bird and Wildlife Sanctuary, JMF: Jefferson Memorial Forest, LRR: private property in Harrison County, MOC: Mosquito Creek Glades, POC: Post-Oak Cedar Nature Reserve & Harrison-Crawford State Forest, and RHR: Rabbit Hash Ridge,



CHAPTER II

ABIOTIC FACTORS, NOT MYCORRHIZAL ASSOCIATIONS, PREDICT SIZE AND ABUNDANCE OF THE INVASIVE GRASS *MICROSTEGIUM VIMINEUM*.[1]

SUMMARY

Both abiotic and biotic factors can influence distributions of invasive plant species and it is important to understand how such factors contribute to invasion success in order to develop successful management strategies. *Microstegium vimineum*, known as stiltgrass, is an invasive annual grass in US Eastern deciduous forests which can outcompete native understory species, decrease diversity, and prevent the regeneration of native trees. *Microstegium* is also known to form associations with arbuscular mycorrhizal fungi (AMF), but research has yet to demonstrate whether this association has a role in *Microstegium* invasion and dominance over native vegetation. We conducted a field survey in invaded and uninvaded habitats across six sites near Louisville, KY, USA to explore the relative importance of biotic versus abiotic factors in predicting *Microstegium* abundance and size. Canopy openness was the strongest predictor of *Microstegium* abundance, followed by soil moisture. Soil nitrogen was the strongest predictor of *Microstegium* tiller height. Surprisingly, AMF extraradical hyphal

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[1] Koenig, K. & Emery, S. M. 2021. Abiotic factors, not mycorrhizal associations, predict size and abundance of the invasive grass *Microstegium vimineum*. The Journal of the Torrey Botanical Society 148(4):294-307. https://doi.org/10.3159/TORREY-D-20-00049.1.

abundance and root colonization were not significant predictors of *Microstegium* abundance nor size. In terms of abiotic factors, our results confirm previous studies that have demonstrated that *Microstegium* grows best in areas with high light and high soil moisture; however, our analysis provides further insight by assessing the relative importance of each of these factors for *Microstegium* invasion.

INTRODUCTION

Non-native plant invasions can reduce native diversity, modify ecosystem processes, and change physical characteristics of native environments and habitats (Mack et al. 2000). Both abiotic and biotic factors can influence distributions of invasive species. For example, many invasive plants, such as *Lonicera japonica* (Japanese honeysuckle) take advantage of disturbances to establish and spread from light gaps in forests (Lemke et al. 2011). Other invasive plants establish in high-nutrient soils where they can outcompete native species (Daehler 2003). Biotic interactions, such as novel associations with arbuscular mycorrhizal fungi (AMF), can also facilitate plant invasions (i.e. “enhanced mutualism hypothesis) (Reinhart and Callaway 2006, Bunn et al. 2015). Still other research has shown that some non-native plants can reduce the abundance of soil biota such as AMF, thus indirectly suppressing native vegetation (Vogelsang and Bever 2009). It is important to understand how both local environmental conditions and associations with soil biota can influence the abundance and growth of invasive plants to best predict and control their spread.

Microstegium vimineum (Trin.) A. Camus (stiltgrass or browntop, hereafter, *Microstegium*) is an invasive annual grass in deciduous and other forests of the eastern

US. *Microstegium* originates from Asia (Droste et al. 2010), having first been documented in Tennessee in the early 1900s as dried shipping material (Fairbrothers and Gray 1972). Since introduction, the species has spread and is now present in more than 23 eastern US states, including Kentucky and Indiana (Ziska et al. 2015). The grass often creates dense beds on the forest floor, outcompeting native understory species (Flory and Clay 2009), decreasing native diversity, and preventing the regeneration of native tree seedlings (Oswalt et al. 2007). As a C4 warm-season grass, *Microstegium* is an unusual forest invader, showing preference for high light and high moisture conditions (Droste et al. 2010, Huebner 2010), but also demonstrating shade tolerance (Horton and Neufield 1998, Wilson et al. 2015). This species can survive and reproduce in as little as 2-8% full sun by increasing leaf production, modifying tiller height, and altering photosynthetic responses (Cheplick 2005, Huebner 2011, Cheplick 2015). Adaptation to shade has allowed the species to spread into forested areas, especially along streambanks, trails, roadsides, and canopy gaps (Redman 1995) via water, human activity, animal activity, recreational vehicles, mowing, and forestry (Novy et al. 2013).

Microstegium is also known to be mycorrhizal, and the association is correlated with an increase in biomass production (Lee et al. 2014), however, research is lacking as to the role mycorrhizal associations play in *Microstegium* invasion. *Microstegium* may take advantage of beneficial AMF in forest soils to compensate for shallow root systems and to outcompete native plants, which has been shown in other systems such as tallgrass prairie (Hartnett and Wilson 1999). This association may be particularly important in dry, shady habitats where AMF may help *Microstegium* overcome abiotic stresses (Winter et al. 1982, Horton and Neufield 1998, Touchette and Romanello 2010, Warren et al. 2011a, Lee et al. 2012).

In this study, we explored the relative importance of biotic versus abiotic factors in predicting *Microstegium* abundance and size (specifically tiller height, as shoot size is positively correlated with reproductive mass (Cheplick and Fox 2011)). We expected to confirm previous studies that show that *Microstegium* is most abundant in high-light, high-soil moisture conditions (Cheplick 2010, Droste et al. 2010, Flory and Clay 2010, Touchette and Romanello 2010, Warren et al. 2011a), and that *Microstegium* would grow taller in sunny areas with open forest canopies (Cheplick 2005). We also predicted that biotic factors (soil AMF abundance and root colonization of *Microstegium*) would have positive significant relationships with plant abundance and size (Reinhart and Callaway 2006).

MATERIALS AND METHODS

Survey sites

We conducted a field survey during June 2019 in six sites located near Louisville, KY, USA (Appendix I Supplementary Figure 16). These six sites contained similar tree species and understory composition. Common species found in these sites included *Acer* spp. (maple), *Quercus* spp. (oak), *Carya* spp. (hickory), *Asimina triloba* (pawpaw), *Parthenocissus quinquefolia* (Virginia creeper), *Muhlenbergia schreberi* (nimblewill), and *Impatiens capensis* (jewelweed). Sites varied in size (66-6530 ha, average size = 1608 ha), underlying geology and soil characteristics, and the extent of *Microstegium* invasion (10-70%) (Appendix I Supplementary Table 7). No management of

Microstegium had been performed in the sampled areas of this study, though some sites actively managed *Microstegium* and other invasive species in adjacent areas (Appendix I Supplementary Table 7).

Data collection

A 300m x 300m grid was overlaid onto a map of each field site using ArcGIS mapping software (ArcMap 10.6.1), and sixteen sampling points, 100m apart, were identified at the corners and intersections of the grid (Fig. 2). We arranged the plots to fit within the property limits, avoid sensitive areas or protected species, and to avoid plots being placed on roads or trails, in streams, within steep ravines, or in other inaccessible areas. We were therefore unable to control for slope or aspect as we were limited in the available placement of the grid within the site. This method was used instead of a paired-plot design, in order to take advantage of the gradient of invasion across sites, and as not all sites had areas where native and corresponding nonnative plots could be placed within a reasonable distance.

In the field, the pre-identified points were located using a Garmin GPS unit, and a 1m x 1m quadrat was placed on the ground for sample collection. Due to difficult terrain and local flooding, only fifteen points from Jefferson Memorial Forest and eight points from Bernheim Research Forest were able to be sampled, resulting in a total of 87 points, 38 of which contained *Microstegium.*

At each sample point, we recorded soil moisture (General DSMM500 meter) in four locations within the sampling quadrat and averaged these values. We measured percent canopy cover using a convex model densiometer. We also collected eight 2.5cm x 10cm soil cores in each quadrat, which were combined for nutrient analyses. Soil chemical analyses were conducted by the University of Kentucky Soil Testing Laboratory (Lexington, KY), for % total N, P, and pH. We visually estimated the percent cover of *Microstegium* in each sampling quadrat. *Microstegium* height was obtained by measuring four tillers per quadrat: the tallest, the shortest, and two haphazardly selected. We then averaged these values to give one value.

We collected an additional four 2.5cm x 10cm soil cores per quadrat, combined them, and transported them to the lab to measure extraradical hyphal (*ERH*) abundance (cm g^{-1} soil) using standard methods (McGonigle et al. 1990, Emery and Rudgers 2014). To characterize AMF abundance in host roots, ten *Microstegium* tillers with attached roots were collected from each sample quadrat and taken to the lab to quantify % AMF root colonization based on hyphal presence in stained root samples using standard methods (McGonigle et al. 1990, Emery and Rudgers 2014). Roots were cleared by soaking in 10% KOH for 1.5 hours in an 80°C water bath and then stained using Trypan Blue. Ten 1cm samples per plot were affixed to slides and analyzed for presence or absence of AMF.

Statistical Analysis

We used regression analysis in the form of model averaging to evaluate the relative importance of abiotic and biotic factors in predicting *Microstegium* cover and height (Burnham and Anderson 1998). For this approach, statistical models of all possible combinations of predictor variables were compared using weighted Akaike Information Criterion (AIC). The most suitable models included variables with average coefficients significantly different from zero. Importance of each response variable was determined through the summed model weights (∑*wi*) across all models, which gives a value between 0 and 1, with higher values indicating the inclusion of the variable in the best-fit models as determined by the weighted AIC. The significance level and effect size of each predictor variable was determined from the model average coefficients. Model averaging analysis was performed in R software, version 3.6.2 (R Development Core Team 2019), using packages MuMIn, dplyr, and relaimpo.

We used canopy cover, soil moisture, soil pH, soil nitrogen, and soil phosphorous as abiotic predictor variables and soil ERH as a biotic predictor variable for all analyses. To predict *Microstegium* tiller height, we only used data from sampling points where *Microstegium* was present (n=38) and so could include both ERH and AMF root colonization as biotic predictors. A pairwise correlation analysis confirmed no significant relationships between predictor variables.

RESULTS

The model that best predicted *Microstegium* cover (AIC = 206.50, n=87, Appendix I Supplementary Table 8) included four predictor variables: % canopy cover, % soil moisture, soil phosphorous, and soil nitrogen ($F_{4,82}$=16.95, p<0.001, R^2=0.453, Appendix I Supplementary Table 9A-D). Percent canopy cover and soil moisture were the strongest predictors (importance values, IV, of 1.00), with increased *Microstegium* cover associated with low canopy cover and high soil moisture (Fig. 3). Soil phosphorous (IV=0.899) was also positively associated with *Microstegium* cover, while soil nitrogen

(IV=0.681) was negatively associated with *Microstegium* cover. ERH and pH were excluded from the best fit model.

The model that best predicted *Microstegium* tiller height (AIC = 103.3, n=38, Appendix I Supplementary Table 8) included three predictor variables: soil nitrogen, AMF % root colonization of *Microstegium*, and soil ERH abundance ($F_{3,34}$=3.397, p=0.029, R^2=0.231, Appendix I Supplementary Table 9A-D). Soil nitrogen was the only significant predictor (IV=0.935), with larger plants found in areas with higher soil nitrogen (Fig. 4). Canopy cover, soil moisture, soil phosphorous, and soil pH were excluded from the best fit model.

DISCUSSION

Results from this study indicate that associations with AMF are less important than abiotic factors in predicting abundance and tiller height of *Microstegium*. While we can confirm that *Microstegium* does associate with AMF (Lee at al. 2014), neither AMF abundance in soils nor roots were associated with *Microstegium* cover or size, indicating that *Microstegium* does not alter AMF hyphal abundance in the soil, nor preferentially invades areas with increased AMF hyphal abundance. These results do not support the Enhanced Mutualism Hypothesis (Reinhart and Callaway 2006), indicating instead that abiotic factors, especially light, water, and nitrogen availability are more important influences on *Microstegium* growth and abundance in this system. As one caveat, we were only able to look at AMF hyphal abundance in soils and roots in this study, which may not reflect community measures of AMF diversity or functioning (Chagnon et al. 2013). *Microstegium* is known to alter bacterial community composition in the soil

(Kourtev et al. 2002, Fraterrigo et al. 2011), so it is possible that *Microstegium* could also alter AMF diversity or community composition without altering AMF hyphal abundance.

In terms of abiotic factors, our results confirm previous studies that have demonstrated that *Microstegium* grows best in areas with high light and high soil moisture (Redman 1995, Claridge and Franklin 2002, Cheplick 2005, 2010, Droste et al. 2010, Touchette and Romanello 2010, Flory et al. 2011, Warren et al. 2011b, Cheplick 2015). Our analyses provide further insight by being able to address the relative importance of these abiotic factors for *Microstegium* invasions. Our results indicate canopy cover is the most significant predictor of *Microstegium* presence, where higher levels of canopy cover allow less light to penetrate to the understory, restricting *Microstegium* distribution. Soil moisture was the secondary predictor, showing a positive relationship, which also concurs with previous studies (Fairbrothers and Gray 1972, Ziska et al. 2015).

Other abiotic factors were less important than light and soil moisture in predicting *Microstegium* abundance and height. Soil phosphorous and nitrogen were minor predictors of *Microstegium* abundance, though in opposite directions. *Microstegium* is known to inhabit soils with high levels of phosphorus (McGrath and Binkley 2009), and so this may partially explain why AMF associations are not critical for invasion, as AMF may not be as beneficial in areas where phosphorous is less limiting (Lee et al. 2014). *Microstegium* presence is also associated with low soil nitrogen, which may be due to increased uptake (Fraterrigo et al. 2011), slower internal nitrogen cycling compared to native species (DeMeester and Richet 2010), and preferential uptake of nitrate over other forms of N (Ehrenfeld et al. 2001, Fraterrigo et al. 2011, Lee et al. 2012). Despite previous studies showing that *Microstegium* is associated with increased (more basic) soil pH (Kourtev et al. 1998, McGrath and Binkley 2009), pH was not found to be a predictor of *Microstegium* cover in this study. However, our sites differed in underlying bedrock geology (limestone versus sandstone) and had a wide range of soil pH (4.5 to 7.9), indicating *Microstegium* can invade and persist in soils with varying pH levels.

Microstegium height was also strongly influenced by abiotic factors, particularly soil nitrogen. These results are consistent with previous findings of plasticity in *Microstegium* traits across abiotic gradients (Claridge and Franklin 2002, Droste et al. 2010, Lee et al. 2012), specifically the positive correlation between canopy openness and tiller height (Claridge and Franklin 2002, Cole and Weltzin 2004, Leicht et al. 2005, Christen and Matlack 2009), which may allow *Microstegium* to outcompete other species for light. Surprisingly though, we found that soil nitrogen had a much stronger relative effect on tiller height than canopy cover. A few previous studies have reported that added nitrogen increased aboveground biomass and seed production in *Microstegium* (Fraterrigo et al. 2011, Ross et al. 2011), which could indicate that habitats with higher levels of soil nitrogen could be under increased invasion pressure as has been found in other systems (Vasquez et al. 2008).

Conclusions

Results from our work indicate that the role of AMF in *Microstegium* invasion success is limited. While AMF can impact plant community structure (Bever et al. 2010) as well as plant diversity within the community (Vogelsang et al. 2006), novel management options that suppress AMF abundance in the soil to release native C_3

grasses and forbs from competitive inhibition by dominant C_4 grasses (Hartnett and Wilson 1999) are not expected to affect *Microstegium* abundance. Our results suggest that management options for control of *Microstegium* invasion should continue to focus on limiting disturbances in natural areas to avoid increased canopy light gaps (Baiser et al. 2008, Cheplick 2010, Warren et al. 2011b), along with other approaches such as mowing, hand pulling, and grass-specific herbicide (Gibson et al. 2002, Judge et al. 2008, Flory 2010, Ward and Mervosh 2012).

Figure Legends

Figure 2: Example of a 300m survey grid with marked sampling points at the Rabbit Hash Nature Reserve in Harrison County, Indiana.

Figure 3: Coefficients from model averaging predicting percent cover of *Microstegium*. Distance from 0 indicates level of importance, with variables closest to 0 not included in the best fit model. Horizontal bars indicate +/- 1 standard error. All plots (invaded and noninvaded) were used in the analysis (n=87).

Figure 4: Coefficients from model averaging predicting average tiller height of *Microstegium.* Distance from 0 indicates level of importance, with variables closest to 0 not included in the best fit model. Horizontal bars indicate +/- 1 standard error. Only the plots containing *Microstegium* were used in the analysis (n=38).

Figure 2

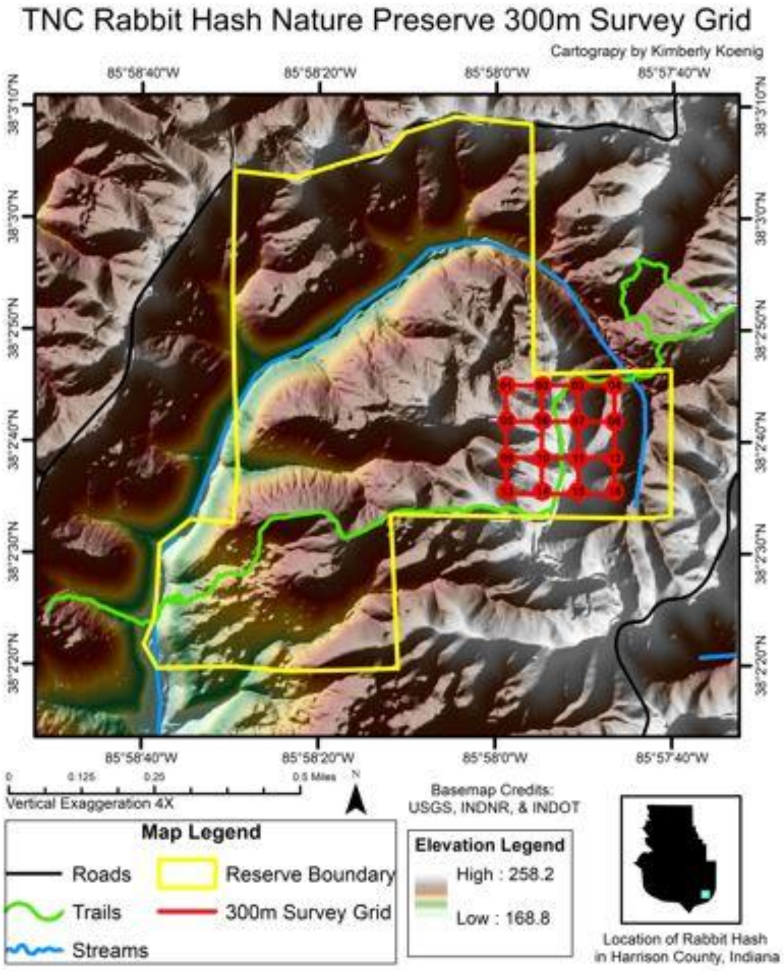
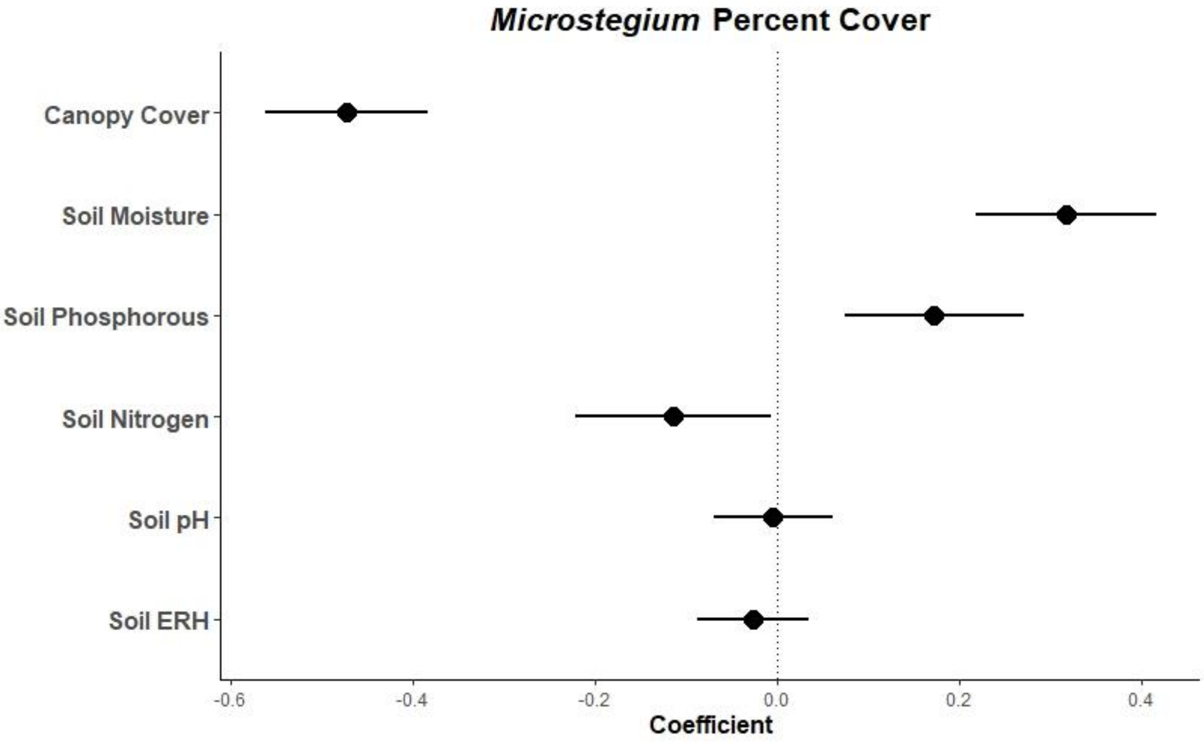
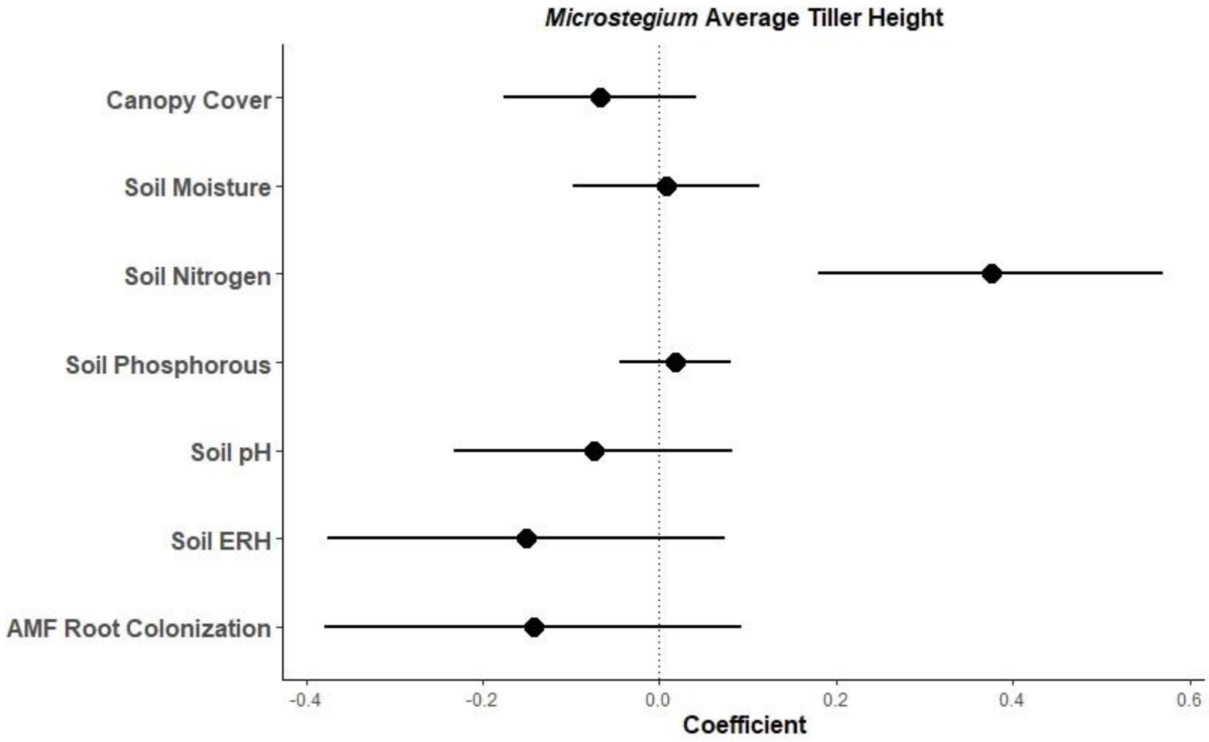

TNC Rabbit Hash Nature Preserve 300m Survey Grid
Cartograpy by Kimberly Koenig
85°58'40"W
85°58'20"W
85°58'0"W
85°57'40"W
38°3'10"N
38°3'0"N
38°2'50"N
38°2'40"N
38°2'30"N
38°2'20"N
0
0.125
0.25
0.5 Miles
N
Vertical Exaggeration 4X
Basemap Credits:
USGS, INDNR, & INDOT
Map Legend
Roads
Reserve Boundary
Trails
300m Survey Grid
Streams
Elevation Legend
High : 258.2
Low : 168.8
Location of Rabbit Hash
in Harrison County, Indiana

Figure 3



Microstegium Percent Cover
Canopy Cover
Soil Moisture
Soil Phosphorous
Soil Nitrogen
Soil pH
Soil ERH
-0.6
-0.4
-0.2
0.0
0.2
0.4
Coefficient

Figure 4



Microstegium Average Tiller Height
Canopy Cover
Soil Moisture
Soil Nitrogen
Soil Phosphorous
Soil pH
Soil ERH
AMF Root Colonization
-0.4
-0.2
0.0
0.2
0.4
0.6
Coefficient

CHAPTER III

SEVERAL NATIVE FOREST PLANTS HAVE REDUCED GERMINATION AND GROWTH IN SOILS ASSOCIATED WITH *MICROSTEGIUM VIMINEUM* INVASION.

SUMMARY

Altered soil conditions are potential mechanisms contributing to the success of invasive plant species. *Microstegium vimineum* is a non-native grass that invades deciduous forests and woodlands in the eastern United States. Its occurrence is associated with changes in soil nutrients and the soil microbial community. Such changes potentially inhibit germination and growth of native species while facilitating growth of *Microstegium*. The goal of this study was to determine whether native plant species differed in germination and growth when grown in soil from *Microstegium* invaded vs. non-invaded woodland areas. While soil type did not alter woody species germination rates, we found increased post-germination mortality for native woody species and decreased germination for native grasses in invaded soils. However, one high-value species, *Quercus rubra*, showed increased aboveground growth in soils associated with *Microstegium* invasion. *Microstegium* seeds also tended to have higher germination success in soils associated with invasion. Overall, the results of this study indicate that altered soil conditions may facilitate *Microstegium* invasion, while simultaneously contributing to population declines of native species.

INTRODUCTION

Plants interact with soils in complex ways, including by influencing soil conditions that enhance their own growth via changes to soil nutrients, soil bacterial communities, or mycorrhizal communities (Bennett and Klironomos 2018; Ehrenfeld and Scott 2001, Wolfe and Klironomos 2005, Reinhart and Callaway 2006, Bever et al 2012). Such changes to the soil can be particularly important for non-native species, where altered soil conditions can increase plant invasion success (Ehrenfeld et al. 2001), suppress native plant growth (Suding et al. 2013), and modify plant community structure (van der Putten et al. 2013, Mack and Bever 2014, Bauer et al. 2015, Lekberg et al. 2018), independent of direct competitive effects between plants.

Altered soil conditions are known to be an important aspect of plant invasion success in deciduous forest ecosystems. For example, *Lonicera maackii* (Amur honeysuckle) is a widespread invader of eastern forests of the United States, and its leaf litter contains a distinct microbial community from native communities which may alter nutrients available to native plants (Arthur et al. 2012). The invasion of the biennial forb, *Alliaria petiolata* (garlic mustard) in forests depresses metabolic function of the soil microbial community (Price and Blair 2019), increases putative fungal pathogens in soils (Anthony et al. 2017, Duchesneau et al. 2017), depresses ectomycorrhizal abundance in the soil (Anthony et al. 2017), and can shift ectomycorrhizal composition (Duchesneau et al. 2017), all which may reduce forest tree regeneration. Other invasive forest plant species, such as *Ranunculus ficaria* and *Celastrus orbiculatus* have been found to suppress germination and growth of native species via changes to soil chemistry (Cipollini and Bohrer 2016).

Microstegium vimineum, (hereafter, *Microstegium*) commonly known as stiltgrass, is an invasive annual grass which forms dense populations in eastern US forest and woodland systems (Novy et al 2013), prohibiting the regeneration of native herbaceous and woody species via various mechanisms (Oswalt et al. 2007, Flory and Clay 2009). For example, *Microstegium* invasion depresses growth of native grass species by reducing access to light (Leicht et al. 2005). *Microstegium* also reduces native tree regeneration through light competition (Aronson and Handel 2011) and increased fire intensity (Flory et al. 2015). It is unlikely that this species competes well with other plants for soil-based resources because of its shallow root system (Flory and Clay 2010a, Flory and Clay 2010b), however changes to soil properties associated with *Microstegium* invasion may indirectly impact native plant species.

Microstegium invasion is associated with changes to multiple soil properties, such as increased soil pH (Kourtev et al. 1998, Ehrenfeld et al. 2001, Cole and Weltzin 2004, McGrath and Blinkley 2009, Ross et al. 2011, Warren et al. 2011), altered carbon cycling (Strickland et al. 2011), altered soil microbial community composition (Kourtev et al. 2002), and decreased soil microbial biomass (Fraterrigo et al. 2011). *Microstegium* invasion can additionally alter nitrogen cycling and nitrate availability (Lee et al. 2012) through acceleration of microbial decomposer activity (Craig and Fraterrigo 2017). Moreover, *Microstegium* promotes the growth of ammonium oxidizing chemoautotrophs (Shannon-Firestone et al. 2015) leading to the production of nitrate (Rippel et al. 2020), which is the favored form of nitrogen for uptake (Ehrenfeld et al. 2001). By altering soils in these ways, *Microstegium* potentially enhances its own success via positive feedbacks

(Ehrenfeld et al. 2001) or by inhibiting the growth of native species (Oswalt et al. 2007, Aronson and Handel 2011, Johnson et al. 2015).

We performed a series of lab experiments using field-collected soil from invaded and uninvaded habitats to determine if soils associated with *Microstegium* invasion can suppress native species germination success and growth in the absence of direct competition. We further determined whether *Microstegium*-associated soils simultaneously stimulate *Microstegium* growth. We predicted that *Microstegium* would have higher germination success in invaded soils, characteristic of positive plant-soil feedbacks typical in plant invasions (Lee et al. 2012). By contrast, we expected that native species would have higher germination success and growth in non-invaded soils, as shown in other studies (Suding et al. 2013). Finally, we explored whether differences in mycorrhizal root colonization could be the mechanism explaining any differences in plant growth, as these organisms have been implicated in other studies of soil conditioning (Anthony et al. 2017, Duchesneau et al. 2017), and are known to be important tree symbionts in eastern deciduous forests (Dickie et al. 2002, Ambriz et al. 2010, Craig et al. 2019, Kumar et al. 2020).

MATERIALS AND METHODS

Tree Germination and Growth Experimental Design

A single-factor experiment was used to assess the effect of soil conditioning on the germination success and aboveground and belowground biomass of 12 native woody species commonly found co-occurring with *Microstegium*: *Acer saccharum* (sugar maple), *Asimina triloba* (pawpaw), *Carya cordiformis* (bitternut hickory), *Carya ovata* (shagbark hickory), *Cercis canadensis* (Eastern redbud), *Diospyros virginiana* (persimmon), *Fraxinus americana* (white ash), *Fraxinus pennsylvanica* (green ash), *Liriodendron tulipifera* (tulip poplar), *Quercus alba* (white oak), *Quercus rubra* (red oak), and *Quercus velutina* (black oak). All tree seeds were purchased from Sheffield's Seed Company (Locke, NY), except *Liriodendron tulipifera* and *Quercus alba* which were collected in Harrison County, Indiana in the fall of 2020. Seeds were cold or warm stratified according to species specifications prior to planting. *Carya cordiformis* and *Fraxinus americana* failed to germinate, and *Quercus velutina* and *Liriodendron tulipifera* had such low germination rates (<0.05%) they were not included in analysis. *Diospyros virginiana* was only included in the germination analysis due to low germination (7%) and seedling die-off prior to harvest.

In 2019, soils were collected from paired invaded and uninvaded field locations in three sites near Louisville, KY (Table 2): University of Louisville's Horner Bird and Wildlife Sanctuary in Crestwood, Kentucky (HWS), the Nature Conservancy's Rabbit Hash Ridge in Harrison County, Indiana (RHR), and a private property in Harrison County, Indiana (LRR). For invaded locations, a 0.5m x 0.5m area with greater than 90% *Microstegium* cover was chosen for soil sampling. *Microstegium* plants were removed from the plot and the top 10cm of soil was collected, as this is where most *Microstegium* roots are found. A similar soil sample was collected from a nearby site (<50 m) with 0% *Microstegium*. Collected soils were sieved (4mm) to remove rocks and organic items (aboveground and belowground biomass, woody debris, live organisms, etc.) and stored at 4°C prior to use in experiments. Subsamples of collected soils were sent to the University of Kentucky Soil Analysis Lab (Lexington, KY) to determine total soil N, P, and pH before experiment initiation (Table 2). Native and invaded soils were not combined across sites to avoid confounding site effects, as recommended by Rinella and Reinhart (2018).

To measure germination and growth of tree species in response to soils associated with *Microstegium* invasion, we filled conical root trainer pots (Deepots, 3.8cm x 21cm for smaller seeds and 6.9cm x 35.5cm for larger seeds, from Stuewe & Sons, Inc., Tangent, OR) 80% full of neutral growth medium (1:1:1 of Bracto Sphagnum Peat Moss, Vigoro Organic Perlite, and Vigoro Organic Vermiculite) and then added field-collected soils as appropriate to fill each pot to 90% capacity. One seed was then added to each pot and covered with neutral growth medium to full pot capacity. Soils from each site (Table 2) were used in ten replicates (total of 30 in invaded soil, 30 in native soil). Pots were kept under grow lights (Gro-Lux Wide Spectrum) on a 14-hour light cycle, maintained at room temperature of 20°C, and kept at consistent saturation by watering twice weekly.

For all pots, seed germination date was recorded, and tree seedlings were grown for 16-24 weeks. After plants showed no increased growth for two weeks, plant height (cm) was measured from the soil surface to the top of the apical meristem, and plants were then harvested (16 weeks for *Fraxinus pennsylvanica,* 17 weeks for *Quercus spp*., 19 weeks for *Carya ovata*, 20 weeks for *Acer saccharum*, and 24 weeks for *Asimina triloba).* Harvested material was separated into aboveground (leaf and stem separate) and belowground biomass, dried at 65°C for at least five days, and then weighed. Any plants that germinated but failed to survive to harvest were recorded as dead.

Tree Species Data Analysis

To evaluate the effect of soil type on germination success, aboveground biomass, and belowground biomass of native tree species, we used a variance-components linear mixed model (estimated using REML and nloptwrap optimizer) with species, soil, and species × soil interactions as fixed effects. The model included site as a random effect. Model 95% confidence intervals (CIs) and p-values were computed using a Wald t-distribution approximation. Germination success, mortality, and average seedling growth were calculated from the 10 technical replicate pots per soil type in each site. This approach ensured robust tests of soil type effects while avoiding pseudo-replication (Hurlbert 1984, Engel et al. 2012, Blainey et al. 2014). Data were analyzed with a Type III analysis of variance (ANOVA) with Satterthwaite's method. We used post-hoc estimated marginal means pairwise comparisons when the species × soil interaction was significant. We used Shapiro-Wilk tests to determine normality. Raw stem height data were normally distributed, whereas percent germination, days to germination, aboveground biomass, belowground biomass, and mortality required square-root or arcsine square-root transformations to meet assumptions of normality. All statistical analyses were performed in R version 4.2.1 (R Development Core Team 2022), with packages lme4 (Bates et al. 2015) and lmerTest (Kuznetsova et al. 2017).

Mycorrhizal Root Analysis

We examined mycorrhizal colonization of roots for the two tree species that showed significant changes in biomass across soil treatments: *Fraxinus pennsylvanica* and *Quercus alba*. These two species differ in their known associations with mycorrhizal fungi, with *Fraxinus* associating with arbuscular mycorrhizal fungi (AMF; Ambriz et al.

2010, Craig et al. 2019), and *Quercus* strongly associating with ectomycorrhizal fungi (EMF; Dickie et al. 2002, Craig et al. 2019, Kumar et al. 2020). We measured percent AMF root colonization of *Fraxinus* based on hyphal presence in stained root samples using standard methods (McGonigle et al. 1990, Emery and Rudgers 2014). In brief, *Fraxinus* root samples were cleared with 10% KOH and stained with Trypan Blue, and 10 1cm root pieces were mounted to slides using PVLG glue. Root pieces were scored at 200x magnification based on presence or absence of fungal hyphae only, as arbuscules were not developed. To determine the effect of soil type on AMF root colonization, we used a variance-components linear mixed model (estimated using REML and nloptwrap optimizer) with soil as a fixed effect, followed by a Type III analysis of variance (ANOVA) with Satterthwaite's method.

Quercus roots were analyzed for the presence of EMF infection through the staining method introduced in Daughtridge et al. (1986) and outlined in Holopainen and Vaittinen (1988). In brief, an 8cm section of dried root from each plant, including all root branches and fine hairs, was stained in a solution of 10% acetic acid containing 0.1% Ponceau S (acid red) and evaluated for EMF infection under 200x magnification. The entire root segment was analyzed and scored based on presence or absence of EMF fungi, as indicated by hyphae of the Hartig net, fungal mantle, or extraradical hyphae.

Grass Germination Assays Design

We conducted a second experiment to assess the effect of soil conditioning on the germination of *Microstegium* and of four native grass species: *Elymus virginicus* (Virginian wild rye), *Elymus hystrix* (bottlebrush), *Panicum anceps* (fall panicum), and

Muhlenbergia schreberi (nimblewill). These native species are known to be present in local reserves where *Microstegium* is also found. *Microstegium* seeds were collected from a private property in Harrison County, Indiana during October of 2018. Seeds of native species were purchased from Roundstone Native Seed (Upton, KY) in 2019.

We used a standard bioassay to compare germination success for *Microstegium* and the four native grass species between invaded and native soils (Dabney et al. 1996; Lou et al. 2016). In brief, a 50cm x 29.2cm sheet of plastic wrap (Glad ClingWrap, Clorox Co., Oakland, CA) was covered with a 38cm x 25.4cm layer of EPA recycled nonperforated paper towel (Georgia Pacific, Atlanta, GA). Seeds were arranged 3cm apart on the towel, which was moistened with 20 ml tap water. Seeds were covered with 100g of field soil from the relevant treatment and moistened with an additional 40mL of water. Another 38cm x 25.4cm layer of paper towel was placed on top of the soil and moistened with 20mL of water. The whole unit was rolled up, and the bottom and the top were folded and secured with a rubber band to maintain internal moisture. Each germination assay unit contained ten seeds, with ten technical replicates for each soil sampling site per native species, resulting in 600 seeds planted per native species (300 in invaded soils and 300 in native soils). We had twice as many *Microstegium* assay units (n=20 per soil sampling site) as we were initially interested in comparing germination rates between two different seed source populations. However, these differences were negligible and so all data from the two different *Microstegium* populations were combined for subsequent analyses.

The germination assay units were stored vertically in a plastic container under ambient room lighting and temperature conditions, except for *Elymus hystrix* and

Panicum anceps which required heat for germination and were placed on seedling heat mats which provided a steady temperature at 5.5-11.1°C above ambient air temperature. The top towel layer was removed to record germination every seven days for 28 days, at which point the final germination was recorded and the trial terminated.

Grass Germination Assays Statistical Analysis

To evaluate the effect of soil type on germination success of native grasses, we used a variance-components linear mixed model (estimated using REML and nloptwrap optimizer) with species, soil, and species × soil interaction as fixed effects. The model included site as a random effect. Model 95% confidence intervals (CIs) and p-values were computed using a Wald t-distribution approximation. Germination success was averaged across the 10 technical replicates per soil type in each site to provide robust tests of soil type effects while avoiding pseudo-replication (Hurlbert 1984, Engel et al. 2012, Blainey et al. 2014). As described above, we used a Type III ANOVA with Satterthwaite’s method to determine the effect of species and soil, and their interaction. Specific differences among treatments were determined with post-hoc pairwise comparisons of marginal means using package emmeans (Length 2022). All germination data were arcsine square root transformed to meet assumptions of normality.

To evaluate the effects of soil type on *Microstegium* germination we also used a variance-components linear mixed model, but with only soil type as the fixed effect. We compared these results with a similar mixed model analysis treating each replicate as independent, due to low power of the most conservative model and ongoing debate about how to treat biological vs. technical replicates in plant-soil feedback studies (Hurlbert 1984, Engel et al. 2012, Blainey et al. 2014).

RESULTS

Tree Germination and Mortality

While species differed in their germination rates, time to germination, and mortality rates, we found no significant effect of soil type nor of any species × soil interaction on germination (Table 3, Appendix II Supplementary Table 10). We did find weak evidence ($p<0.1$; Muff et al. 2022) for soil type effects on tree seedling mortality, with seedlings in invaded soils having 83.6% higher mortality than those in uninvaded soils (Table 3, Fig. 5, Appendix II Supplementary Table 10), and this was consistent across species (i.e., no species x soil interaction).

Tree Seedling Growth

We found strong evidence that tree seedling species varied in their aboveground growth responses across soil types (Table 3, Appendix II Supplementary Table 10). While most species showed no growth response differences across soil types, *Quercus rubra* had 75% more aboveground biomass and was 72% taller in invaded soil types (Fig. 6, Appendix II Supplementary Table 10). We found no soil treatment effects on seedling belowground biomass, even though belowground biomass differed between species (Table 3, Appendix II Supplementary Table 10).

Mycorrhizal Root Colonization

Root colonization by arbuscular mycorrhizal fungi in *Fraxinus pennsylvanica* was generally low in both native (20.0%) and invaded soils (17.3%) and did not differ between soil treatments (F(1)= 0.4495, p=0.510; data not shown). All of the *Quercus* root samples from field soil treatments showed evidence of EMF presence, and so were not analyzed for statistical differences among soil treatments.

Grass Germination

We found weak evidence (p<0.1; Muff et al. 2022) that grass germination varied between soil types, with invaded soils decreasing germination of native grasses by ~53-84% (Table 3, Fig. 7). Germination success also differed among species, but there was no soil × species interaction (Table 3).

Our most conservative analysis of effects of soil type on *Microstegium* germination showed no significant effects (Table 3). However, when replicates were treated as independent, average germination of *Microstegium* was 69% higher in invaded soils than in native soils (Appendix II Supplementary Figure 17).

DISCUSSION

Plant invasion can potentially modify soil conditions to enhance an invader's own growth while inhibiting growth of co-occurring native species (Suding et al. 2013). Here, we show that field-collected soils associated with *Microstegium* invasion tend to promote self-germination while suppressing germination of native grass species. We also found increased mortality for native woody species in *Microstegium* invaded soils, though, counter to our hypotheses, invaded soils promoted growth for one native tree species. The results of this study indicate that *Microstegium* invasion alters plant communities in deciduous forests via multiple mechanisms.

Microstegium potentially contributes to tree seedling mortality by altering soil microbial communities (Kourtev et al. 2002), including changes in soil pathogenic fungal loads or composition. While examining the community of soil pathogens was beyond the scope of our study, *Microstegium* invasion could potentially enhance the load of plant pathogens commonly associated with our focal species. For example, damping off, a fatal disease caused by various pathogenic fungi, is a common cause of seedling mortality in *Quercus spp.* and *Fraxinus spp.* (Yamazaki et al. 2009), as is root rot in *Quercus spp.* (Yamazaki et al. 2009). Additionally, *Acer spp., Cercis spp.*, and *Fraxinus spp.* are susceptible to *Verticillium* wilt (Aljawasim and Vincelli 2015), and *Carya ovata* is susceptible to *Phytopthora* root rot (Brazee et al. 2017). *Microstegium* litter can alter microbial community structure and function (Elgersma et al. 2012) and can promote pathogen infections on native grasses such as *Elymus hystrix* (Benitez et al. 2022); therefore, increases in litter cover associated with *Microstegium* invasion can lead to soil conditions that promote woody plant pathogens.

One oak species (*Quercus rubra*) exhibited increased growth in response to *Microstegium* invaded soils, specifically increased aboveground biomass and stem height. This result concurs with other research showing that *Microstegium* increases native plant productivity in the context of shifting pathogen effects during direct plant competition (Shannon et al. 2012). Another common forest invader, *Alliaria petiolata*, also increases the growth of native species due to altered function and composition of the soil microbial community (Price and Blair 2019). Whether shifts in the microbial community associated with *Microstegium* (e.g., Kourtev et al. 2002) benefit woody conspecifics remains unknown. The results of this study suggest that changes to mycorrhizal fungi are unlikely to explain growth benefits to native plants; moreover, they support our previous work showing no differences in mycorrhizal activity in invaded and uninvaded forest sites (Koenig and Emery 2021).

The suppressive effects of *Microstegium* invaded soils on native grass species germination could result from multiple abiotic and biotic mechanisms. A key potential abiotic effect is altered soil nitrogen and carbon (Kourtev et al. 1998, Ehrenfeld et al. 2001, Strickland et al. 2011, Warren et al. 2011, Kramer et al. 2012). Potential biotic effects include allelopathy (Pisula and Meiners 2010, Corbett and Morrison 2012) and altered soil microbial communities (Fraterrigo et al. 2011, Kramer et al. 2012, North and Torzilli 2017). Soil analysis data from this study suggest that biotic factors are driving the observed differences. Specifically, it is likely that increased abundance of at least one pathogen reduced the germination success of native plants. *Microstegium* hosts a foliar fungal pathogen, *Bipolaris gigantea,* which also infects native grasses like *Elymus virginicus* (Kendig et al. 2021, Benitez et al. 2022) and can suppress germination (Kleczewski et al. 2012). *Microstegium* may also have long acting allelopathic effects in soils, although the evidence is mixed (Barden 1987, Cole and Weltzin 2004, Cipollini and Bohrer 2016), with some inhibitory allelopathic effects on seed germination and growth observed in the lab (Pisula and Meiners 2010, Corbett and Morrison 2012). Regardless, more work is needed to identify specific mechanisms that suppress germination of native species in *Microstegium* invaded soils.

Whereas soils associated with *Microstegium* decreased germination and survival of both herbaceous and woody native species, *Microstegium* itself tended to grow better in those soils. Positive responses for invasive plants in a non-native range have been found for many species, including *Prunus serotina* (black cherry) (Reinhart et al. 2003), *Centaurea maculosa* (spotted knapweed) (Callaway et al. 2004), *Alliaria petiolata* (garlic mustard), *Euphorbia esula* (leafy spurge), *Cirsium arvense* (Canada thistle), and *Lythrum salicaria* (purple loosestrife) (Klironomos 2002). The mechanisms driving the positive responses in invaded soils in this study are uncertain. In other studies, *Microstegium* invasion was associated with altered nitrogen cycling and increased nitrification rates (Lee et al. 2012), which can break dormancy and promote seed germination in some systems, especially for pioneer species (Auchmoody 1979, Giba et al. 2003, Pérez-Fernández et al. 2006, Luna and Morena 2009, Zhong et al. 2019). Changes in soil pH can also alter germination in some species (Pérez-Fernández et al. 2006, Shoemaker and Carlson 1990), However, the soils used in this study did not differ in any predicable way for these properties. Thus, changes to soil microbes are a more likely explanation (e.g., Gallery et al. 2007, Li et al. 2017, Guo et al. 2021). As a caveat, while initial invasions may benefit from reduced soil pathogens (e.g., van der Putten 2002); with time *Microstegium* can eventually accumulate high pathogen loads which may help with long-term management (Flory and Clay 2013).

Conclusions

Microstegium invasion can suppress forest regeneration through mechanisms such as direct light or soil resource competition, increased litter depth, increased deer herbivory, and changes to fire intensity (Kourtev et al. 1998, Leicht at al. 2005, Baiser et al. 2008, Flory and Clay 2010a, Flory and Clay 2010b, Aronson and Handel 2011, Johnson et al. 2015, Averill et al. 2016). The results of this study suggest that changes to the soil associated with *Microstegium* invasion may further contribute to declines of native species. Increased tree seedling mortality will contribute to slow forest regeneration, while decreased herbaceous species germination could alter small mammal, bird, and reptile communities that depend on herbaceous species in forests for shelter and food resources (Durner and Gates 1993, McCollin 1998, Howe et al. 2002, Carfagno et al. 2006) However, one high-value species, *Quercus rubra*, was able to thrive in soils associated with *Microstegium* invasion. Understanding the many mechanisms by which *Microstegium* impacts the regeneration and growth of both woody and herbaceous native species is critical to understanding forest dynamics and management of timber resources.

Table 2: Soil characteristics for LRR (private property in Harrison County, IN), RHR (the Nature Conservancy's Rabbit Hash Nature Reserve in Harrison County, IN), and HWS (the University of Louisville's Horner Wildlife and Bird Sanctuary in Oldham County, KY).

Site	**Latitude/Longitude**	**Soil**	**Total Soil N**	**P** (kg/ha)	**pH**
LRR	38.0322 -85.9882	Native	0.20%	49.32	6.3
LRR	38.0324 -85.9881	Invaded	0.18%	24.66	6.1
RHR	38.0482 -85.9655	Native	0.15%	7.85	5.9
RHR	38.0481 -85.9585	Invaded	0.18%	20.18	5
HWS	38.3434 -85.5268	Native	0.24%	6.73	5.9
HWS	38.3428 -85.5286	Invaded	0.20%	125.54	7.5

Table 3. Results of mixed models examining the effects of plant species and soil treatments on germination and growth responses. Significant effects are in bold (p<0.10).

	Native Grasses:			*Microstegium:*		
	% Germination			% Germination		
	df	F†	p	df	F†	p
Plant Species	3	**6.37****	0.0048	N/A	N/A	N/A
Soil Type	1	**3.67***	0.0733	1	3.65	0.1962
Species * Soil	3	1.32	0.3033	N/A	N/A	N/A
Site (Random)	1	<0.0001	1.0000	1	0.0370	0.0840

	Trees:			Trees:			Trees:		
	% Germination			Days to Germination			Seedling Mortality		
	df	F†	p	df	F†	p	df	F†	p
Plant Species	8	**20.13*****	<0.001	8	**54.36*****	<0.001	8	**22.30*****	<0.001
Soil Type	1	0.88	0.3543	1	0.67	0.4193	1	**3.80***	0.0616
Species * Soil	8	0.72	0.6692	7	1.05	0.4206	7	1.32	0.2789
Site (Random)	1	0.0006	0.8269	1	0.0138	0.3929	1	0.0024	0.4579

	Trees:			Trees:			Trees:		
	Aboveground Biomass			Belowground Biomass			Stem Height		
	df	F†	p	df	F†	p	df	F†	p
Plant Species	6	**200.94*****	<0.001	6	**401.18*****	<0.001	6	**57.10*****	<0.001
Soil Type	1	**11.91****	0.0019	1	0.01	0.9317	1	**11.81****	0.0020
Species * Soil	6	**3.44***	0.0123	6	1.81	0.1386	6	**4.99****	0.0016
Site (Random)	1	0.0000	1.0000	1	0.0014	0.1425	1	0.0000	1.0000

* indicates significance at 0.1, ** indicates significance at 0.01, and *** indicates significance at 0.001; † : Variance provided for Site (random effect).

Figure Legends

Figure 5: Seedling mortality across soil treatments (medians and quartiles). P-value obtained from mixed-model ANOVA.

Figure 6: *Quercus rubra* (red oak) average (medians and quartiles) aboveground biomass and stem height at harvest. P-values obtained from post-hoc comparison of means.

Figure 7: Germination (medians and quartiles) of native grass species across soil treatments. P-value obtained from mixed-model ANOVA.

Figure 5

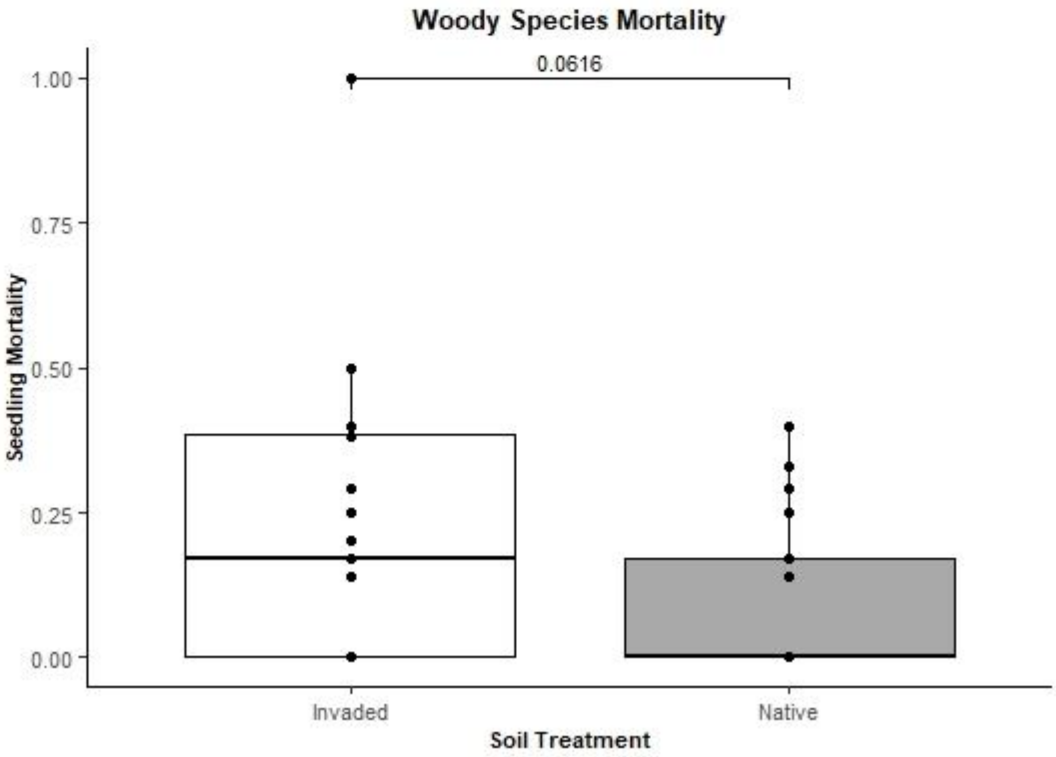
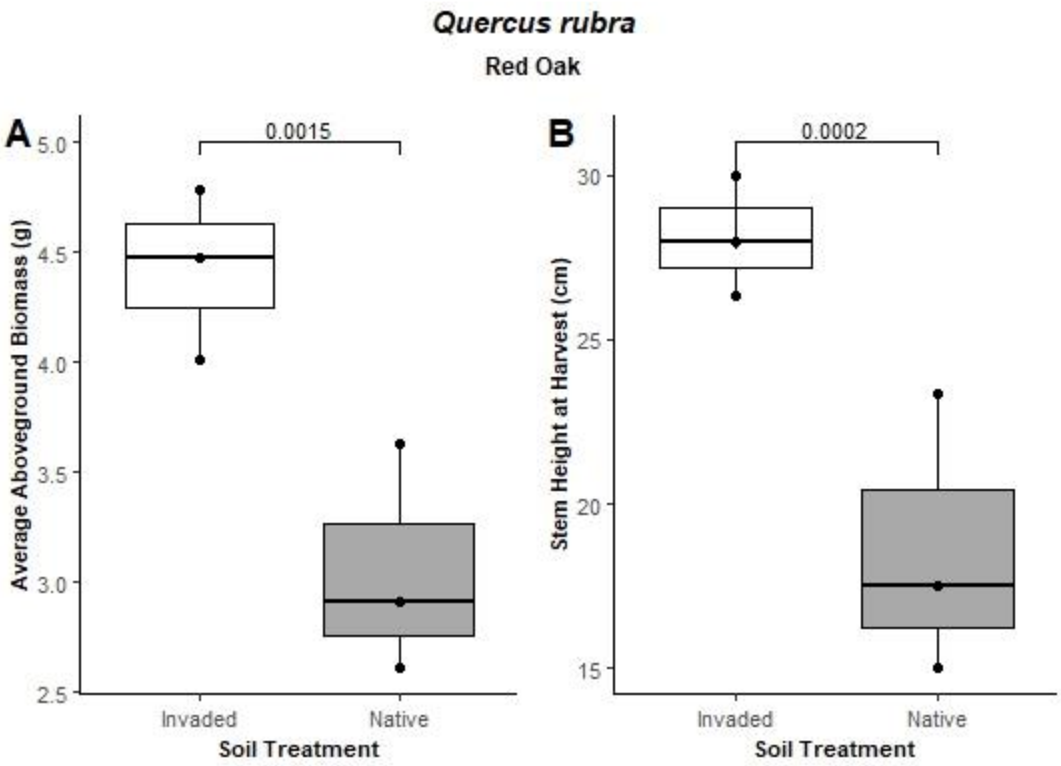
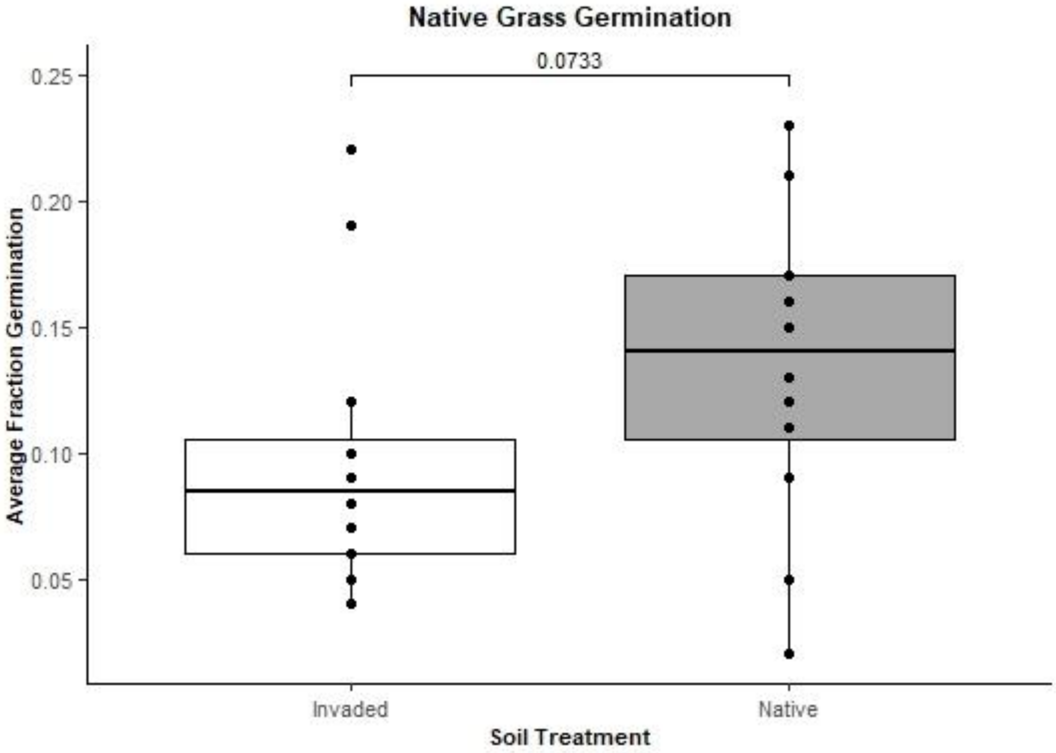

Woody Species Mortality
0.0616
1.00
0.75
0.50
0.25
0.00
Seedling Mortality
Invaded
Native
Soil Treatment

Figure 6



Quercus rubra
Red Oak
A
0.0015
5.0
4.5
4.0
3.5
3.0
2.5
Average Aboveground Biomass (g)
Invaded
Native
Soil Treatment
B
0.0002
30
25
20
15
Stem Height at Harvest (cm)
Invaded
Native
Soil Treatment

Figure 7



Native Grass Germination
0.0733
0.25
0.20
0.15
0.10
0.05
Average Fraction Germination
Invaded
Native
Soil Treatment

CHAPTER IV

MICROSTEGIUM VIMINEUM HABITAT SUITABILITY ANALYSIS IN THE KENTUCKIANA REGION USING GEOGRAPHIC INFORMATION SYSTEM (GIS) MODELING.[2]

SUMMARY

Microstegium vimineum or stiltgrass is a novel invader widely considered one of the most significant threats to oak-hickory woodlands in the eastern United States. Due to the insubstantial weight and buoyancy of seeds, *Microstegium* can disperse not only via overland dispersal routes but also via sheet flow, flooding, and stream channels, creating rapid progression of existing populations and greater likelihood of introduction to new areas. Using the knowledge of *Microstegium* habitat from previous research and field mapping of current populations, we identified and assessed environmental variables for use in habitat suitability modeling. We found canopy openness, distance from roads, and distance from streams to be the primary drivers of *Microstegium* presence. We then applied a GIS-based habitat suitability analysis across seven nature reserves in Kentuckiana as a potential tool for land management. Our pilot study developed a model which can potentially guide managers of natural areas to decide where best to focus

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[2] Koenig, K. & Day, C.A. 2023. *Microstegium vimineum* habitat suitability analysis in the Kentuckiana region using Geographic Information System (GIS) modeling. Southeastern Geographer 63(2):183-201.

mitigation and prevention of potential *Microstegium* invasion and population expansion when resources are limited.

INTRODUCTION

Biotic invaders are species introduced to a novel region in which they establish, thrive, spread, and persevere to the detriment of the native environment. Exotic invasions can reduce native diversity, modify ecosystem processes, and change the physical characteristics of native environments and habitats (Mack et al. 2000). One such invader, *Microstegium vimineum,* commonly known as stiltgrass, is an invasive annual grass originating from Asia (Droste et al. 2010), which is now present in twenty-three eastern states, including Kentucky and Indiana (Ziska et al. 2015). *Microstegium* grows into dense populations (Novy et al. 2013) through the production of lateral tillers and the generation of copious amounts of seeds (Gibson et al. 2002), which can disperse via water, human activity, recreational vehicles, mowing, and forestry (Novy et al. 2013).

Microstegium often dominates habitats with high light availability (Cole and Weltzin 2004, Cheplick 2005, Warren et al. 2011, Flory et al. 2017), high soil moisture (Webster et al. 2008, Touchette and Romanello 2010), and where understory litter has been removed or disturbed (Barden 1987, Oswalt et al. 2007, Marshall and Buckley 2008). Although an annual grass with a C4 photosynthetic pathway, *Microstegium* can penetrate the forest interior and survive in diffuse light conditions typically dominated by species with C3 photosynthetic pathways (Christen and Matlack 2009, Warren et al. 2011). *Microstegium* populations may expand and disperse along linear areas of disturbance, such as road banks, animal trails, fire trails, logging roads, utility right of ways, and stream channels (Redman 1995), as well as via sheet flow in wetlands and floodplains (Fairbrothers and Gray 1972).

Microstegium seeds are buoyant, and can disperse in overland water flow via surface runoff and flooding (Cole and Weltzin 2004, Cheplick 2010, Warren et al. 2011), or along stream courses (Miller and Matlack 2010), wildlife paths (Huebner 2010), diversion channels (Christen and Matlack 2009), and roadsides (Rauschert et al. 2010). Total seed movement is positively correlated with precipitation (Tekiela and Barney 2013), with populations moving downslope two to four times faster than upslope (Schramm and Ehrenfeld 2012) and larger patches occurring along roadsides and within wet meadows (Nord et al. 2010).

In the 1980s, the U.S. Fish and Wildlife Service created Habitat Suitability Indices, or ranking systems to determine the most likely areas of habitation by a specific species, which were utilized to create habitat suitability models (U.S. Fish and Wildlife Service 1981). When Geographic Information System (GIS) software became available, map models were created using known species habitat requirements and available environmental data. GIS habitat suitability modeling has since been used as a management tool for various species. For example, habitat was assessed for beavers in Austria (Maringer and Slotta-Bachmayr 2006) using topographical (slope and elevation), hydrological (water bodies, stream networks, and water levels), and vegetative inputs (land use and forest cover), and for crested ibis in China (Li et al. 2002) using vegetation type, elevation, water networks, and human disturbance. Habitat suitability modeling was also used to inform calcareous grassland restoration in the United Kingdom (Burnside et al. 2002) using topographical and vegetative community data, and to predict plant community distribution in Greece (Vogiatzakis and Griffiths 2006) using vegetative responses to altitude, slope, and landform. Additionally, habitat suitability modeling has been utilized to predict areas of potential invasion by *Mimosa pigra* in Vietnam (Le et al. 2019), by considering habitat variables such as elevation, slope, aspect, solar insolation, topographic wetness index (TWI), distance from the nearest road, and a Normalized Difference Vegetation Index (NDVI).

Due to the ability of *Microstegium* seeds to travel along linear disturbance routes and via buoyancy in water, the goal of this study was to develop a pilot habitat suitability analysis for *Microstegium*. We determined the most important *Microstegium* habitat requirements through a literature review and statistical analysis of habitat variables for current populations mapped in the field. For managers of Eastern deciduous forest reserves with limited resources, habitat suitability research could potentially direct limited resources to slow *Microstegium* spread and minimize the impact of further species invasion.

MATERIALS AND METHODS

Field Sites

All sites contain Eastern deciduous oak-hickory forest systems and are located within the Interior Low Plateau level III ecoregion. The three field sites in Harrison County, Indiana, are in the Mitchell Plain level IV ecoregion, characterized by karst topography and leached loess soils (Woods et al. 2002a). The Mosquito Creek Nature Preserve contains forest glades with species specific to limestone bedrock. The Brock-Sampson Nature Reserve in Floyd County, Indiana, is within the adjacent level IV ecoregion of the Norman Upland, characterized by knobs and hills, narrow valleys, high order stream channels, and silty loam soils (Woods et al. 2002a). This site also contains forest glades with species specific to sandstone bedrock. The three sites in Kentucky all lie within the Outer Bluegrass level IV ecoregion characterized by variable local relief and mostly composed of Upper Ordovician limestone and shale (Woods et al. 2002b). Louisville and the surrounding areas experience a continental climate characterized by hot, muggy summers and cold, wet winters. Annual precipitation falls just under 115 cm and annual average temperatures range from 9.2–14.5 °C (Fig. 9).

Field Data Collection

Each field site was visited between 9–28 July, 2020, and surveyed for the presence of and percent cover of *Microstegium*, which typically germinates in March or April, becoming established by early June. ArcGIS Collector was used with the assistance of a BadElf Pro GPS to collect data in the field. At each property, *Microstegium* populations were mapped wherever observed within accessible areas of each property, which excluded steep terrain, densely forested areas, and areas where park managers prohibited access. GPS locations were mapped along the edge of each population and in a grid pattern within the population, if the patch was large enough to do so. Each location point was evaluated for percent cover in a 0.25 x 0.25 m area quadrat, estimated by eye to the nearest 10 percent (Table 4). Point density per site was dependent on the level of *Microstegium* invasion and the accessibility of areas within the site. Point layers were exported to ArcMap 10.6.1 and ArcGIS Pro 2.9 to be used for habitat suitability analysis.

Determination of Habitat Variables

A literature review revealed the following habitat variables as correlated with *Microstegium* presence: open canopy or high light conditions (Cole and Weltzin 2004, Cheplick 2005, Warren et al. 2011, Flory et al. 2017, Koenig and Emery 2021), soil moisture (Redman 1995, Webster et al. 2008, Touchette and Romanello 2010), elevation (Schramm and Ehrenfeld 2012, Culpepper et al. 2018), and proximity to roads (Christen and Matlack 2009, Cheplick 2010, Rauschert et al. 2010), trails (Gibson et al. 2002, Cheplick 2010), and streams (Christen and Matlack 2009, Miller and Matlack 2010).

GIS Data Acquisition and Manipulation

A GIS data layer was acquired for each of the determined habitat variables (Table 2), with topographic wetness index (TWI) serving as a proxy for soil moisture to incorporate potential soil moisture availability and topography (Hengl and Reuter 2009) (Table 5). Reserve boundaries for the Hayswood Nature Reserve and the Brock-Sampson Nature Reserve were obtained through INDNR Managed Lands files acquired through IndianaMap, and the boundaries for Rabbit Hash Ridge and the Mosquito Creek Nature Preserve were provided directly by The Nature Conservancy. Boundaries for Broad Run Park and Jefferson Memorial Forest were obtained from LOJIC via Louisville Metro Parks data, and a boundary file for the Horner Bird and Wildlife Sanctuary was created using georeferencing.

Each input layer was projected to the NAD 1983 UTM 16N coordinate system for uniformity and clipped to the respective reserve boundary, as applicable (not every variable was present at every reserve, Appendix III Supplementary Table 11). Canopy cover and elevation rasters were utilized as acquired, while proximity to roads, streams, and trails was derived by creating a Euclidean distance raster from the acquired line features. Topographic wetness index was derived utilizing methods outlined by Moeslund et al. (2013) to create an index raster (Table 5).

Statistical Analysis of Habitat Variables

A Pearson's *r* correlation matrix was completed for all habitat variables and *Microstegium* percent cover, and model averaging was used to determine the relationship between *Microstegium* percent cover and the habitat variables (Burnham and Anderson 1998, Dormann et al. 2018). All data analysis was completed in the software R, version 4.1.2 (R Development Core Team 2020), with Pearson's correlation computed using the Hmisc package and model averaging using MuMin. For model averaging, the data was scaled to improve the stability of the models and the accuracy of the estimated parameters for each variable. The model averaging analysis ran regression models with different combinations of variables and computed a weight for each model based on parsimony with the data. Weights increase with model fit, and the best possible model was determined by analyzing comparisons of weighted AIC (Aikake Information Criterion) outputs (Burnham and Anderson 1998). The best model included variables with average coefficients differing significantly from zero. The importance of each response variable was expressed as $\sum wi$, or the sum of model weights across all models, which returns a value between 0 and 1. Values closer to one indicate the variable is more likely to be included in the best-fit model.

Habitat Suitability Analysis

In ArcGIS, each variable layer was reclassified into three levels indicating ideal, acceptable, or unsuitable habitat areas (Appendix III Supplementary Table 11). Classification was decided based on a literature review of the known parameters of *Microstegium* habitat and for simplicity assumed a linear response omitting other potential response functions. Canopy cover was considered ideal (3) at 0-25 percent (open sun or minimal shade), acceptable (2) at 25-75 percent, and unsuitable (1) at above 75 percent (mostly shade cover) (Appendix III Supplementary Table 11). Proximity to roads, streams, and trails were classified according to distance from the line feature, with 0-10 m ideal (3), 10-20 m acceptable (2), and 20+ meters unsuitable (1) (Appendix III Supplementary Table 11). TWI was classified according to the statistics for the layer, with one standard deviation above and below the mean classified as ideal (3) soil moisture levels, drier areas classified as acceptable (2), and continuously saturated areas classified as unsuitable (1) (Appendix III Supplementary Table 11).

The reclassified layers for each site were added together to produce a habitat suitability map, first with each layer assuming the same level of importance (Fig. 10). The reclassified layers were then weighted according to the absolute value of the coefficient produced by the statistical model averaging and were again summed to produce a second habitat suitability map. The output maps for the non-weighted and weighted analyses were then compared to determine any differences between methods.

RESULTS

Statistical Analysis of Habitat Variables

A Pearson's *r* correlation found *Microstegium* percent cover to be significantly associated with canopy openness ($r = 0.17$, $n = 522$, $p < 0.0001$) and topographic wetness index ($r = -0.15$, $n = 522$, $p = 0.0008$) (Table 6), confirming the inclusion of these two habitat variables in the habitat suitability analysis. The model which best predicted *Microstegium* cover (AIC = 5053.7, $F_{3,518} = 8.674$, $p < 0.001$, $n = 544$) included three predictor variables: canopy openness ($p < 0.001$), distance from roads ($p < 0.001$), and distance from streams ($p = 0.0006$) (Appendix III Supplementary Table 12). All three variables included in the best-fit models had importance values, IV, of 1.00. *Microstegium* cover was directly related to canopy openness ($r = 6.939$), and inversely related with distance to roads ($r = -6.844$) and streams ($r = -4.762$) (Fig. 11, Appendix III Supplementary Table 12)

Elevation was not utilized in the habitat suitability analysis due to a significant association with all other habitat variables and no association with *Microstegium* (Table 6). Topographic wetness index and canopy openness were confirmed as correlated to *Microstegium* presence through Pearson's r correlation (Table 6) and canopy openness, distance to roads, and distance to streams were confirmed as predictors of *Microstegium* percent cover through model averaging analysis. Distance to trails was not confirmed as a habitat variable by either statistical analysis but was included in the habitat suitability analysis due to reports from the literature and the inclusion of the two other linear disturbance pathways.

Habitat Suitability Analysis

Habitat suitability analysis generated two output maps, the first with habitat variables not weighted and the second with habitat variables weighted according to the absolute value of the coefficients acquired through statistical model averaging. Summing the reclassified layers produced a range of areas from high habitat suitability to low habitat suitability, with higher values indicating the presence of multiple habitat variables and lower values indicating the presence of fewer habitat variables at that location.
The first output was not weighted, with each habitat variable assuming equal importance (Fig. 12). Outputs ranged from 3 (low habitat suitability for *Microstegium*) to 13 (high habitat suitability for *Microstegium*). Broad Run Park had output values from 5 to 13, the Brock-Sampson Nature Reserve had values of 3 to 9, the Hayswood Nature Reserve had values of 5 to 13, the Horner Bird and Wildlife Sanctuary had values of 5 to 12, Jefferson Memorial Forest had values of 4 to 12, the Mosquito Creek Nature Reserve had values of 3 to 8, and Rabbit Hash Ridge had values of 4 to 11.

The second habitat suitability analysis weighted variables according to the absolute value of the coefficient produced through statistical model averaging (Fig. 13). Outputs ranged from 12.7 (low habitat suitability for *Microstegium*) to 59.9 (high habitat suitability for *Microstegium*). Broad Run Park had a range of output values from 20.9 to 59.9, the Brock-Sampson Nature Reserve had values of 12.7 to 38.7, the Hayswood Nature Reserve had values of 20.9 to 59.9, the Horner Bird and Wildlife Sanctuary had values of 20.9 to 46.2, Jefferson Memorial Forest had values of 19.5 to 38.5, the Mosquito Creek Nature Reserve had values of 12.7 to 31.2, and Rabbit Hash Ridge had values of 14.1 to 39.5.

DISCUSSION

Canopy openness, or light availability, was confirmed as the strongest predictor of *Microstegium* cover, reinforcing previous findings (Koenig and Emery 2021). Warm season grasses are unusual within forested areas, as most do not support C4 photosynthesis in low light conditions or are outcompeted by C3 species better adapted to shade (Sage and Sultmanis 2016). As a C4-warm season grass, *Microstegium* is an unusual forest invader. Although increased growth and seed production are seen in open canopy conditions (Droste et al. 2010), *Microstegium* demonstrates shade tolerance through photosynthetic induction responses and stomatal closures to prevent water loss in photosynthetically constrained conditions (Horton and Neufeld 1998), increasing invasion success.

Distance to roads and streams are additionally confirmed as predictors of *Microstegium* cover, with denser populations in decreased proximity. Roads and streams offer linear dispersal routes along disturbance pathways, with increased light availability, soil moisture, and soil nutrient availability (Redman 1995, Tickner et al. 2001, Warren et al. 2011, Ziska et al. 2015). Stream banks in particular offer prime habitat with reduced competition and increased nutrient availability through erosion and deposition (Tickner et al. 2001). Seeds of invasive plants such as *Solidago*, *Aster*, *Helianthus*, *Impatiens glandulifera.* and *Fatsia japonica* have migrated via central European river channels (Ellenberg 1988, as cited in Tickner et al. 2001), and in Germany the Bree River was shown to be an effective dispersal agent for invasive wind-dispersed tree species through urban greenways (Saumel and Kowarik 2010).

Surprisingly, topographic wetness index (TWI) was found to have an inverse relationship with *Microstegium* cover. TWI was used as a proxy to incorporate both soil water holding capacity and topographical elevation, as soil moisture was unable to be recorded directly in the field. TWI is limited to predictions and does not incorporate actual levels of rainfall. As previous findings have shown a positive correlation with soil moisture levels (Koenig and Emery 2021), level of precipitation and seasonal flooding may still impact *Microstegium* population expansion. In the Kentuckiana region, *Microstegium* produces and releases seeds in October, which can be transported via seasonal spring flood events to settle in new locations where they may germinate or remain dormant yet viable for three to five years (Barden 1987) until conditions improve. For example, the floodplain of the Brock-Sampson Nature Reserve has been inundated due to the rise of the nearby Ohio River, most notably during February flood events (U. S. Geological Survey, 2022). The importance of the flood-pulse concept in transportation of seeds has been documented by Boedeltje et al. (2004), with highest diaspore numbers occurring in channel waters during elevated autumn and winter discharge.

Habitat suitability analysis showed similar outputs across methods, with linear dispersal paths as the highest rated areas across sites. Weighted variables produced a greater range of suitability scores, with more low suitability areas than the output with variables assuming equal importance. Differences between the outputs were driven by canopy openness, distance to roads, and distance to streams, which had the highest absolute value coefficients from the statistical model averaging (Appendix III Supplementary Table 12). Since the weighted output displays heightened contrast, the areas of high habitat suitability are easier to discern than on the non-weighted output.

Therefore, the weighted method may provide a more valuable resource for management of natural areas.

While not confirmed through subsequent field mapping, this pilot tool can potentially provide valuable assessment information for property managers to focus limited resources both within the site and between sites. For example, The Nature Conservancy manages two field sites included in the study. Mosquito Creek, overall, had lower levels of habitat suitability than Rabbit Hash Ridge, indicating which reserve would benefit most from increased mitigation efforts if resources do not enable the same level of management on both properties.

One limitation to this pilot study was the limited access within the sites, due to challenging terrain and protection of vulnerable areas. Future habitat suitability analyses would benefit from the use of drone imagery to capture the level of invasion across the site and to track population expansion across subsequent years. A pilot drone flight was completed at the University of Louisville's Horner Bird and Wildlife Sanctuary in February of 2021, where *Microstegium* populations had been mapped in July of 2020. The GPS points collected in the field could be used to identify the specific color band where *Microstegium* litter is present, due to the lack of other understory vegetation during the dormant period and expanded to identify all locations on the image where *Microstegium* litter is present. Imaging the same location across several years could determine the level and direction of *Microstegium* expansion and validate the use of habitat suitability analysis as a predictive model.

Conclusions

Microstegium vimineum is widely considered one of the most significant threats to oak-hickory deciduous forests in the eastern United States (Miller and Matlack 2010) and is often listed by land managers as one of the most problematic invasive plants in Kentucky and Indiana woodlands. Current management methods include fire, herbicide, and mowing, however, due to prodigious seed production, plastic responses to novel environments (Cheplick 2005, Cheplick 2008, Cheplick 2015), and human-mediated dispersion (Ziska et al. 2015), management methods have only slowed population expansion. We identified and assessed environmental variables associated with *Microstegium*. We found canopy openness and topographic wetness index to be correlated with *Microstegium* presence, and through statistical model averaging found canopy openness, distance from roads, and distance from streams to be the primary drivers of *Microstegium* presence. We utilized the selected habitat variables to create a GIS-based habitat suitability analysis across seven nature reserves in Kentuckiana, which provided output maps showing high to low areas of suitable habitat for *Microstegium*. Our pilot study showed that habitat suitability analysis could provide a potential tool to help inform land managers with limited resources where best to focus mitigation efforts to prevent further population spread and colonization of *Microstegium*.

Table 4: *Microstegium* populations surveyed at each research site, including area of the property (hectares), number of points collected per property, and the density of points collected at each site (points per hectare).

Site	**Location**	**Area**	**Points**	**Density**
Broad Run Park	Jefferson County, KY	323.39 ha	120	0.37 pts/ha
Jefferson Memorial Forest	Jefferson County, KY	33.59 ha	74	2.20 pts/ha
Horner Bird and Wildlife Sanctuary	Oldham County, KY	72.10 ha	50	0.69 pts/ha
Brock-Sampson Nature Reserve	Floyd County, IN	240.71 ha	46	0.19 pts/ha
Hayswood Nature Reserve	Harrison County, IN	128.87 ha	191	1.48 pts/ha
Rabbit Hash Ridge	Harrison County, IN	134.39 ha	28	0.21 pts/ha
Mosquito Creek Nature Reserve	Harrison County, IN	80.04	13	0.15 pts/ha

Table 5: Acquired GIS data layers for each habitat variable.

Habitat Variable	**GIS Layer**	**Site Data Acquired From**
Elevation (DEM)	USDA 2M LIDAR	Geospatial Data Gateway
Canopy Cover	USFS 2016 Tree Canopy Survey	FS Geodata Clearinghouse
Streams	USGS Flowline	National Hydrograph Database
Roads	USGS Transportation Lines	USGS National Map
Kentucky Trails	USGS Transportation Lines	USGS National Map
Indiana Trails	INDNR Trail Lines	IndianaMAP
Topographical Wetness Index	Derived using USDA 2M LIDAR	Geospatial Data Gateway

Table 6: Pearson's *r* correlation matrix for all habitat variables and *Microstegium* percent cover: ELE = elevation (DEM), TWI = topographic wetness index, CAN = canopy openness, TRL = distance to trails, RD = distance to roads, STR = distance to stream channels, MV = *Microstegium vimineum* percent cover. Significance indicated as: * = 0.05, ** = 0.01, *** = 0.001.

	ELE	TWI	CAN	TRL	RD	STR	MV
ELE	----						
TWI	-0.16***	----					
CAN	0.41***	-0.30***	----				
TRL	0.32***	-0.08	0.08	----			
RD	-0.13**	0.18***	-0.11*	0.07	----		
STR	0.38***	-0.05	0.25***	0.13**	-0.29***	----	
MV	-0.01	-0.15***	0.17***	-0.08	-0.05	-0.04	----

Figure Legends

Figure 8: Location of seven field sites in the Kentuckiana region. Hayswood Nature Reserve, Mosquito Creek Nature Reserve, and Rabbit Hash Ridge in Harrison County, Indiana, Brock-Sampson Nature Reserve in Floyd County, Indiana, Jefferson Memorial Forest and Broad Run Park in Jefferson County, Kentucky, and Horner Bird and Wildlife Sanctuary in Oldham County, Kentucky.

Figure 9: Monthly mean precipitation (cm) and temperatures (°C) for Louisville, Kentucky, over a 30-year period from 1981 to 2010, as obtained from the Kentucky Climate Center. The bars indicate precipitation, the solid line is the average monthly temperature, and the dotted lines are the average high and low temperatures.

Figure 10: Example of input habitat layers and output habitat suitability analysis for Broad Run Park in Louisville, KY. Habitat variable data layers of topographic wetness index (TWI), canopy cover, Euclidean distance to roads, Euclidean distance to streams, and Euclidean distance to trails reclassified and summed to produce the output habitat suitability analysis.

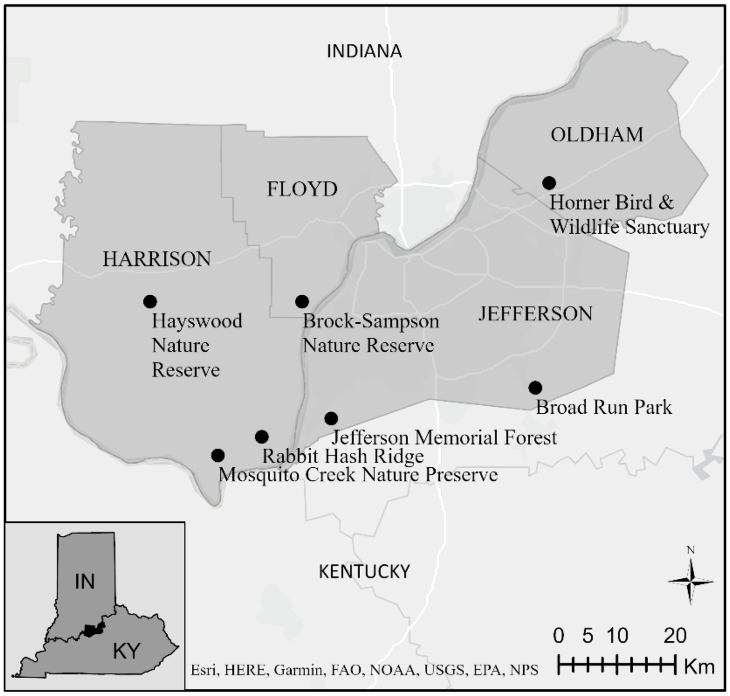
Figure 11: Model averaging coefficients for habitat variables predicting percent cover of *Microstegium*. Distance from 0 indicates level of importance, with variables closest to 0 not included in the best fit model. Horizontal bars indicate +/- 1 standard error.

Figure 12: *Microstegium* habitat suitability analysis output for all seven nature reserves in the Louisville, KY region, with all variables assuming equal importance. Lighter areas indicate high habitat suitability, and darker areas indicate low suitability.

Figure 13: *Microstegium* habitat suitability analysis output for all seven nature reserves in the Louisville, KY region, with all variables weighted according to model averaging coefficient. Lighter areas indicate high habitat suitability, and darker areas indicate low suitability.

Figure 8

Location of Research Sites in Indiana & Kentucky



Esri, HERE, Garmin, FAO, NOAA, USGS, EPA, NPS

Figure 9

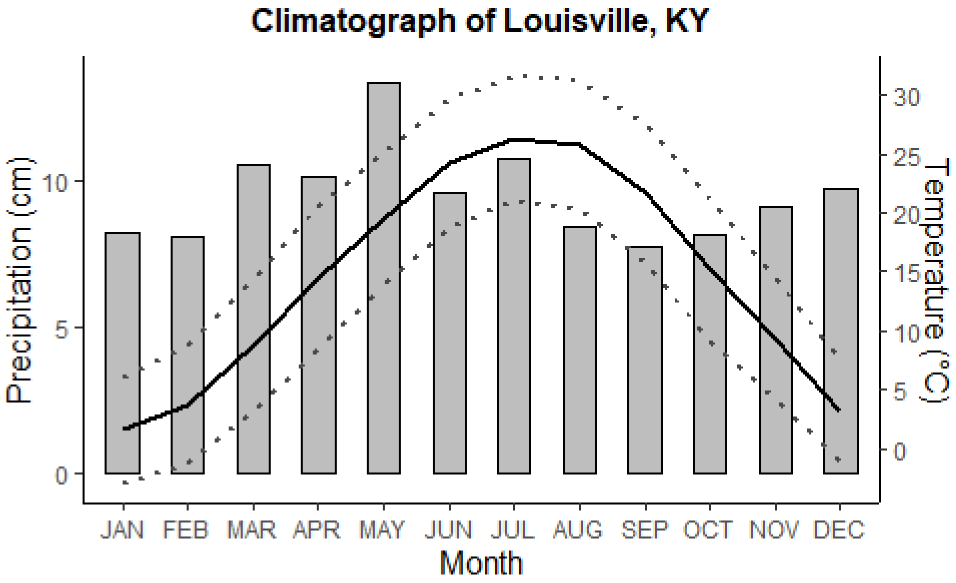

Climatograph of Louisville, KY
Precipitation (cm)
0
5
10
Temperature (°C)
0
5
10
15
20
25
30
JAN
FEB
MAR
APR
MAY
JUN
JUL
AUG
SEP
OCT
NOV
DEC
Month

Figure 10

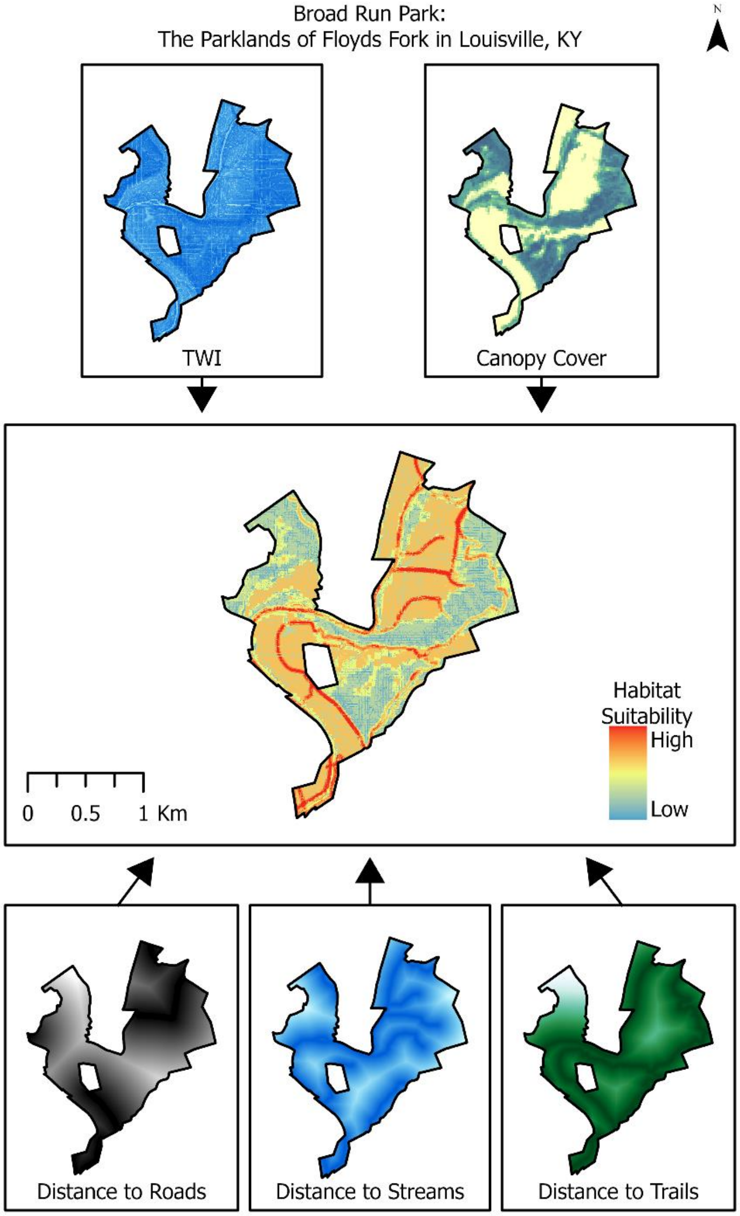

Broad Run Park:
The Parklands of Floyds Fork in Louisville, KY
N
TWI
Canopy Cover
Habitat Suitability
High
Low
0
0.5
1 Km
Distance to Roads
Distance to Streams
Distance to Trails

Figure 11

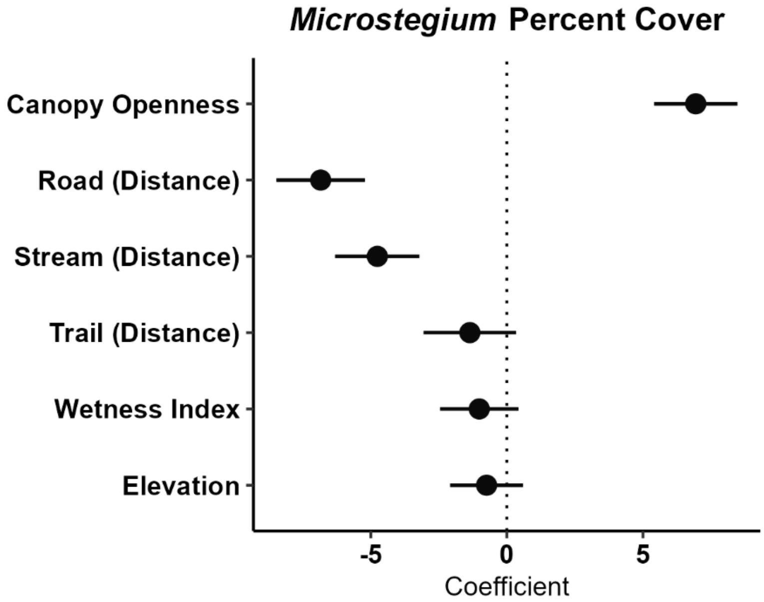

Microstegium Percent Cover
Canopy Openness
Road (Distance)
Stream (Distance)
Trail (Distance)
Wetness Index
Elevation
-5
0
5
Coefficient

Figure 12

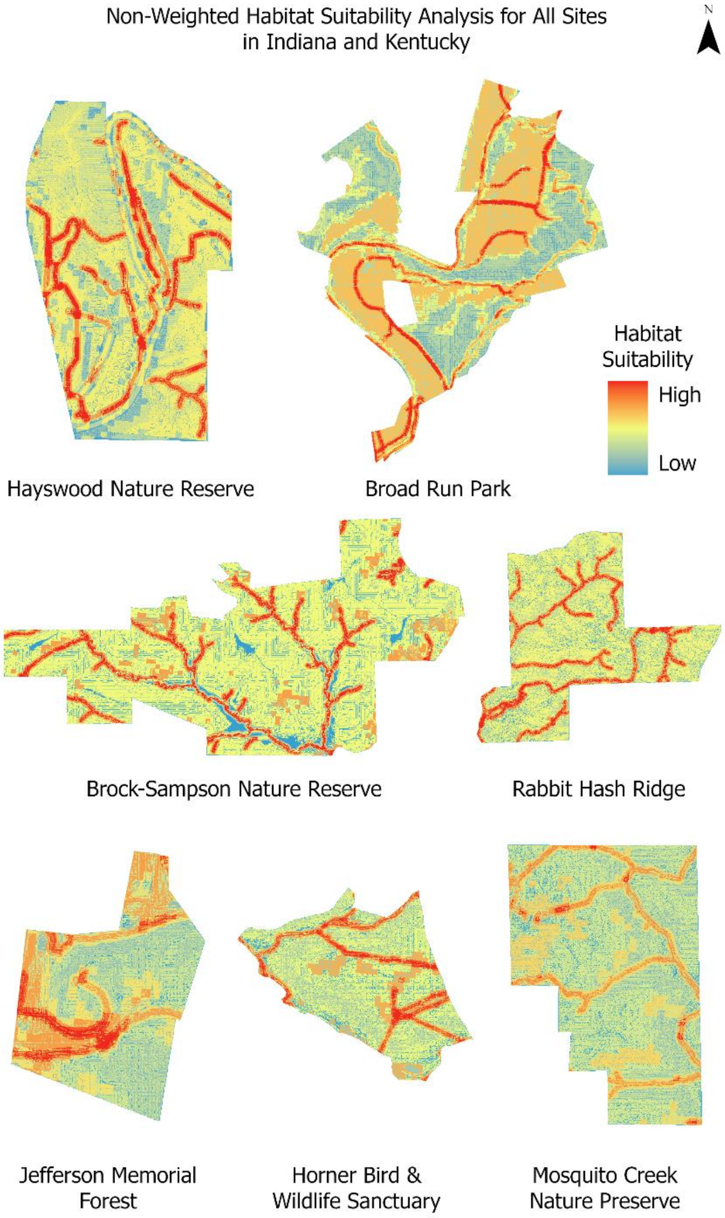
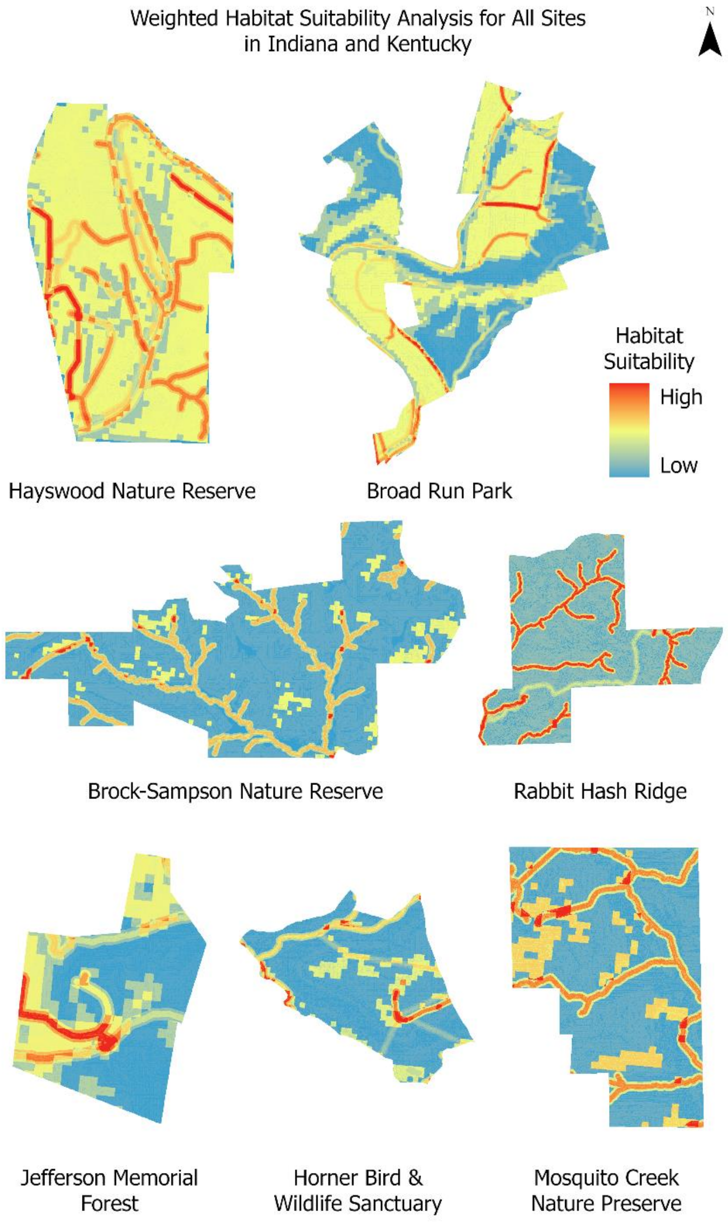

Non-Weighted Habitat Suitability Analysis for All Sites
in Indiana and Kentucky
N
Habitat
Suitability
High
Low
Hayswood Nature Reserve
Broad Run Park
Brock-Sampson Nature Reserve
Rabbit Hash Ridge
Jefferson Memorial
Forest
Horner Bird &
Wildlife Sanctuary
Mosquito Creek
Nature Preserve

Figure 13


Weighted Habitat Suitability Analysis for All Sites
in Indiana and Kentucky
N
Habitat Suitability
High
Low
Hayswood Nature Reserve
Broad Run Park
Brock-Sampson Nature Reserve
Rabbit Hash Ridge
Jefferson Memorial Forest
Horner Bird & Wildlife Sanctuary
Mosquito Creek Nature Preserve

CHAPTER V

SUMMARY AND FUTURE DIRECTIONS

SUMMARY

The results of this dissertation provide insight into the invasion dynamics of *Microstegium* in Eastern deciduous woodlands. I identified several abiotic factors which were strong predictors of *Microstegium* presence. I also demonstrated that soil biota associated with *Microstegium* invasion can impact the germination and growth of *Microstegium* and native species. Finally, I developed a model to predict future *Microstegium* dispersal at the landscape level. Understanding the drivers behind the success of invasive species can inform land management practices at the community and landscape level.

In chapter two, I provided evidence that canopy cover and high soil moisture, followed by soil nitrogen and phosphorous were the strongest predictors of *Microstegium* abundance in the Louisville region. While I confirmed an association between *Microstegium* and arbuscular mycorrhizal fungi, evidenced by the presence of hyphae in roots, this association did not show significant changes across environmental gradients, nor did the concentration of extraradical soil hyphae differ between areas inhabited by *Microstegium* versus native flora. The findings provide no evidence for the enhanced mutualism hypothesis which suggests foreign plant invaders gain an advantage over native species due to a novel association with a beneficial mutualist (Reinhart and

Callaway 2006, Bunn et al. 2015), and instead indicate abiotic factors exert more influence over the ability of *Microstegium* to establish and flourish. These findings also confirm known habitat preferences of *Microstegium* in disturbed areas and along stream or runoff channels.

In chapter three I showed conflicting impacts of soils associated with *Microstegium* invasion on native species. While *Microstegium* exhibited increased germination in soils associated with field populations of this species, germination of native grasses was suppressed and mortality of woody seedlings was enhanced. One native woody species, *Quercus rubra* (red oak), unpredictably showed increased aboveground biomass and stem height at harvest in *Microstegium*-conditioned soils. These results suggest that the lack of regeneration by native woody species observed in the field and documented in common garden experiments (Oswalt et al. 2007, Flory and Clay 2010b, Aronson and Handel 2011, Johnson et al. 2015) is at least partly due to changes in soil conditions, in addition to other mechanisms such as inability for seeds to penetrate the dense litter layer due to the slow rate of decomposition (Kourtev et al. 1998, Ehrenfeld et al. 2001), direct competition with *Microstegium* for light or nutrients (Johnson et al. 2015), or altered fire intensity (Flory et al. 2015).

In chapter four I performed a GIS habitat sustainability analysis for *Microstegium* using known abiotic factors associated to *Microstegium* presence. In seven different preserves, I collected field survey points where *Microstegium* was presently located and recorded the present cover at each point. I then added landscape variable layers to the map, extracted data for the layers at each point, and performed a correlation matrix and model averaging to determine the variable associations with *Microstegium* presence. The variable layers were reclassified into unsuitable – suitable categories and added together to create an output map of habitat suitability. For the second map, the variables were weighted according to the absolute value of the coefficients produced by the model averaging analysis. These output maps provide a potential tool for land managers to direct limited resources to prevent *Microstegium* spread within natural areas.

FUTURE DIRECTIONS

Although I was able to map *Microstegium* population points in the field, topography and physical restrictions at each of the sites limited the amount of data I could collect on foot. To better characterize *Microstegium* populations on the landscape and to validate dispersal models, other means of mapping populations could be utilized. I have already begun this process using drone flights to capture aerial imagery of one of my field sites at Horner Bird and Wildlife Sanctuary in Crestwood, KY. Donald Biddle, Director of University of Louisville's Center for Geographic Information Sciences, completed a drone flight in March of 2021, prior to *Microstegium* germination and native woody species leaf out (Fig. 14). If we can identify *Microstegium* litter as a unique color bandwidth (lighter) as compared to native leaf litter (darker) by comparing aerial imagery with ground collected survey data (Fig. 14), Geographic Information System processing of aerial imagery could be used to identify all areas with this coloration, which would give an indication of *Microstegium* presence across the entire site. If yearly surveys were completed, this technology could map the changes in the *Microstegium* populations at the landscape level and compare these changes to the models of predictive dispersal. The ability to visualize these data without the efforts required to survey populations by foot would greatly assist land managers with prevention and mitigation of *Microstegium*.

One of the most interesting findings of this dissertation was the ability of one native woody species to increase growth in soils associated with *Microstegium* invasion. However, this finding contradicts general field observations of reduced tree seedling success in areas invaded by *Microstegium* (Oswalt et al. 2007, Aronson and Handel 2011, Johnson et al. 2015). Other studies have demonstrated direct competitive interactions between *Microstegium* and native tree species (Baiser et al. 2008, Flory and Clay 2010b, a, Aronson and Handel 2011, Shannon et al. 2012), and indirect negative effects for trees such as altered soil chemistry or fire intensity (Cole and Weltzin 2004, Emery et al. 2011, Fraterrigo et al. 2011, Strickland et al. 2011, Warren et al. 2011b, Lee et al. 2012, Flory et al. 2015). However, forest systems are complex, and include organisms other than native flora which could influence tree regeneration dynamics. For example, by changing habitat cover invasive plants can impact foraging behaviors of small mammals, with cascading effects on native tree seed survival (Mattos and Orrock 2010, Dutra et al. 2011). For example, mice (*Peromyscus leucopus*) were found to alter foraging behaviors in urban areas dominated by the invasive shrub *Lonicera maackii* (honeysuckle), using the shrub as cover to decrease predation risk (Persons and Eason 2016).

To explore potentially similar patterns with *Microstegium*, I started an experiment in April 2021 to evaluate seed foraging behavior in three sites near Louisville, KY. During each season from April 2021-Jan. 2022, I measured seed removal rates of 3-4 different plant species from areas with greater than 90% *Microstegium* cover compared to nearby uninvaded areas. In each area, I placed 10 pans filled with sand and either 6 hickory seeds (*Carya ovata*), 5 oak acorns (*Quercus alba*), 30 Virginia rye seeds (*Elymus virginicus*), or 20 sunflower seeds (*Helianthus* spp.) following methods in Bowers and Breland (1996). Seeds remaining in pans after 48 hours were counted to measure seed predation rates. Preliminary analyses indicate that more seeds are removed from uninvaded areas (Fig.15). Field camera photos and videos showed removal by different animals (mice, raccoons, and squirrels) at different locations. These observations lead to new research questions: Does *Microstegium* presence alter which small mammals are foraging in the area? Since raccoons were often seen consuming the seeds, while squirrels were caching seeds, does *Microstegium* presence alter the abundance of seeds which are consumed vs. cached? And are squirrels caching seeds in both native and invaded areas, or are they removed from the invaded area and cached among native flora due to differences in vegetation and litter abundance? Answering these questions will help determine the mechanism behind the low regeneration rate of native species in *Microstegium*-dominated areas, informing preservation of the remaining Eastern deciduous forest systems.

Figure 14: Imagery from drone flight completed at Horner Bird & Wildlife Sanctuary in March 2021.

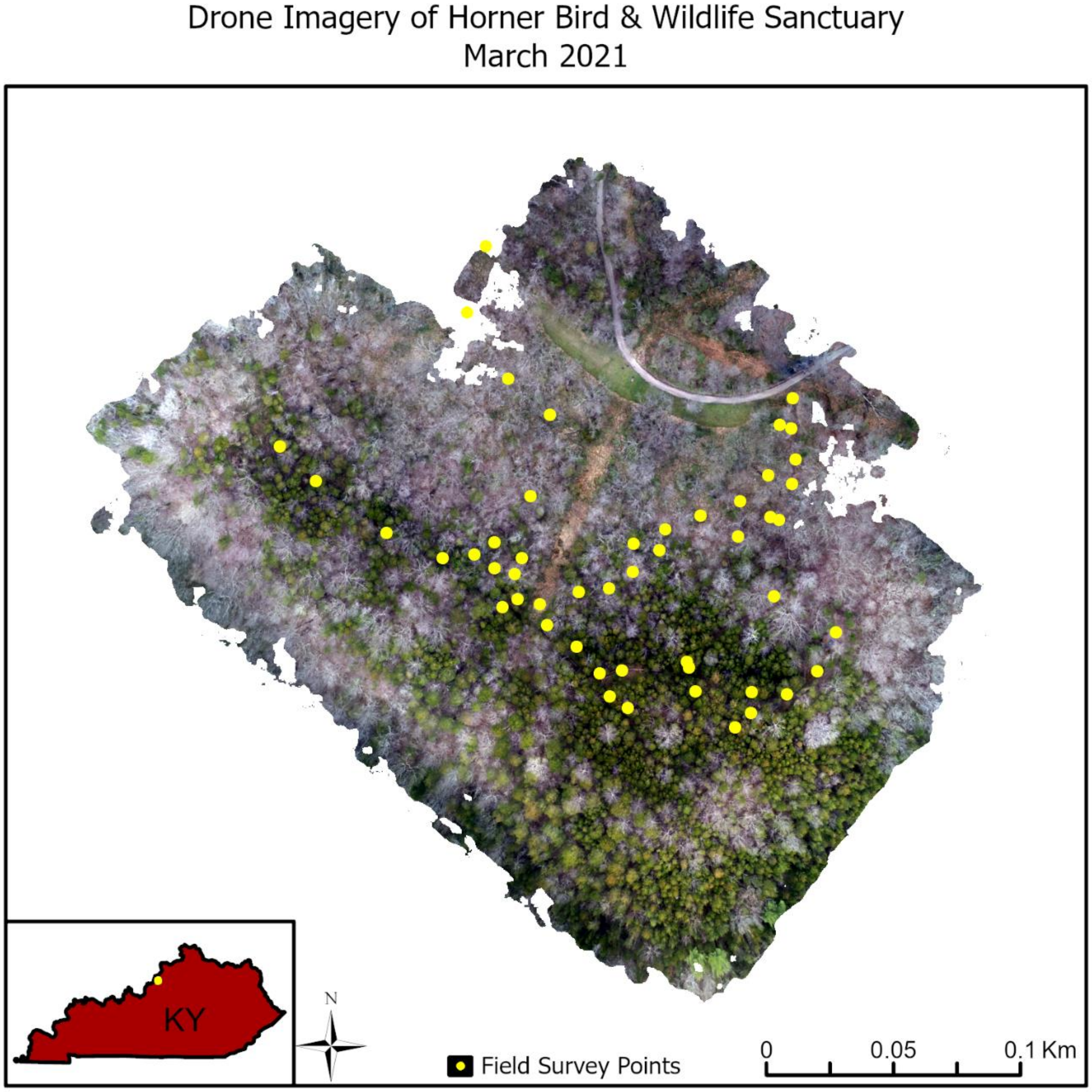
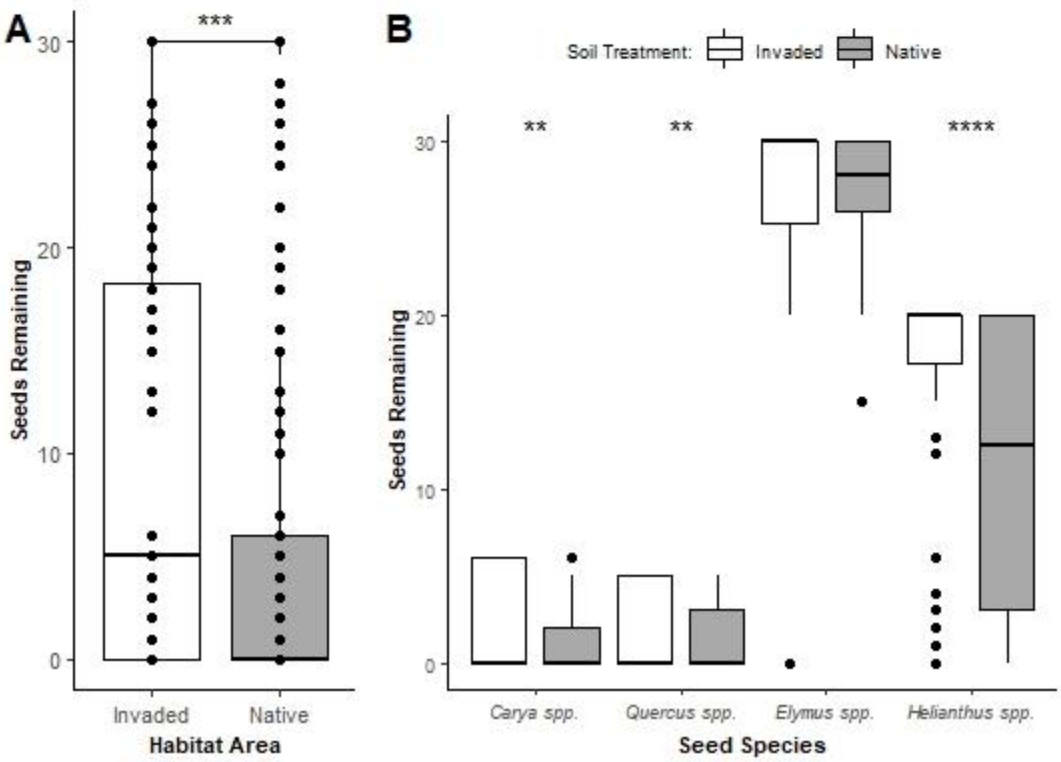


Figure.15: A) Seeds remaining for all species across all sites and seasons. *** indicates significance at 0.001. B) Seeds remaining for each species across all sites and seasons. ** indicates significance at 0.01 and **** indicates significance at 0.0001.



REFERENCES

Adams, S. N., and K. A. M. Engelhardt. 2009. Diversity declines in *Microstegium Vimineum* (Japanese stiltgrass) patches. Biological Conservation 142:1003-1010.

Aljawasim, B., and P. Vincelli. 2015. Evolution of polymerase chain reaction (PCR)-based methods for rapid, accurate detection and monitoring of *Verticillium dahlia* in woody hosts by real-time PCR. Plant Disease 99:866-873.

Ambriz, E., A. Báez-Pérez, J. M. Sánchez-Yáñez, P. Moutoglis, & J. Villegas. 2010. *Fraxinus-Glomus-Pisolithus* symbiosis: plant growth and soil aggregation effects. Pedobiologia 53:369-373.

Anthony, M. A., S. D. Frey, and K. A. Stinson. 2017. Fungal community homogenization, shift in dominant trophic guild, and appearance of novel taxa with biotic invasion. Ecosphere 8:01951.

Arthur, M. A., S. R. Bray, C. R. Kuchle, and R. W. McEwan. 2012. The influence of the invasive shrub, *Lonicera maackii*, on leaf decomposition and microbial community dynamics. Plant Ecology 213:1571-1582.

Aronson, M. F. J., and S. N. Handel. 2011. Deer and invasive plant species suppress forest herbaceous communities and canopy tree regeneration. Natural Areas Journal 31:400-407.

Auchmoody, L. R. 1979. Nitrogen-fertilization stimulates germination of dormant pin Cherry seed. Canadian Journal of Forest Research 9:514-516.

Averill, K. M., D. A. Mortensen, E. A. H. Smithwick, and E. Post. 2016. Deer feeding selectivity for invasive plants. Biological Invasions 18:1247-1263.

Baiser, B., J. L. Lockwood, D. La Puma, and M. F. J. Aronson. 2008. A perfect storm: two ecosystem engineers interact to degrade deciduous forests of New Jersey. Biological Invasions 10:785-795.

Barden, L. S. 1987. Invasion of *Microstegium vimineum* (Poaceae), an exotic, annual, shade tolerant, C-4 grass, into a North Carolina floodplain. American Midland Naturalist 118:40-45.

Bauer, J. T., K. M. L. Mack, & J. D. Bever. 2015. Plant-soil feedbacks as drivers of succession: evidence from remnant and restored tallgrass prairies. Ecosphere 6:158.

Bates, D., M. Mächler, B. Bolker, and S. Walker. 2015. Fitting linear mixes-effects models using lme4. Journal of Statistical Software 67:1-48.

Beasley, R. R., and B. C. McCarthy. 2011. Effects of Microstegium vimineum (Trin.) A. Camus (Japanese stiltgrass) on Native Hardwood Survival and Growth: Implications for Restoration. Natural Areas Journal 31:246-255.

Benitez, L., A. E. Kendig, A. Adhikari, K. Clay, P. F. Harmon, R. D. Holt, E. M. Goss, And S. L. Flory. 2022. Invasive grass litter suppresses a native grass species and promotes disease. Ecosphere 13:e3907.

Bennett, J. A., and J. Klironomos. 2018. Mechanisms of plant-soil feedback: interactions among biotic and abiotic drivers. New Phytologist 222:91-96.

Bever, J. D., I. A. Dickie, E. Facelli, J. M. Facelli, J. Klironomos, M. Moora, M. C. Rillig, W. D. Stock, M. Tibbett, and M. Zobel. 2010. Rooting theories of plant community ecology in microbial interactions. Trends in Ecology & Evolution 25:468-478.

Bever, J. D., T. G. Platt, and E. R. Morton. 2012. Microbial population and community dynamics on plant roots and their feedbacks on plant communities. Annual Review of Microbiology 66:265-283.

Blainey, P., M. Krzywinski, and N. Altman. 2014. Replication. Nature Methods 11:879-880.

Boedeltje, G., J. P. Bakker, A. T. Brinke, J. M. van Groenendael, and M. Soesbergen. 2004. Dispersal phenology of hydrochorous plants in relation to discharge, seed release time and buoyancy of seeds: the flood pulse concept supported. Journal of Ecology 92:786-796.

Brazee, N. J., X. Yang, and C. X. Hong. 2017. *Phytophthora caryae* sp. nov., a new species recovered from streams and rivers in the eastern United States. Plant Pathology 66:805-817.

Bunn, R. A., P. W. Ramsey, and Y. Lekberg. 2015. Do native and invasive plants differ in their interactions with arbuscular mycorrhizal fungi? A meta-analysis. Journal of Ecology 103:1547-1556.

Burnham, K. P., and D. R. Anderson. 1998. Model Selection and Multimodel Inference: A Practical Information-Theoretical Approach. 2d ed. New York: Springer-Verlag.

Burnside, N. G., R. F. Smith, and S. Waite. 2002. Habitat suitability modelling for calcerous grassland restoration on the South Downes, United Kingdom. Journal of Environmental Management 65:209-221.

Callaway, R. M., G. C. Thelen, A. Rodriguez, and W. E. Holben. 2004. Soil biota and exotic plant invasion. Nature 427:731-733.

Campbell, J. 2004. Comparative Ecology of Warm-Season (C4) versus Cool-Season (C3) Grass Species in Kentucky, with Special Reference to Bluegrass Woodlands. Proceedings of the 4th Native Grass Symposium 2005.

Carfagno, G. L., E. J. Heske, and P. J. Weatherhead. 2006. Does mammalian prey abundance explain forest-edge use by snakes? Ecoscience 13:293-297.

Chagnon, P. L., R. L. Bradley, H. Maherali, and J. N. Klironomos. 2013. A trait-based framework to understand life history of mycorrhizal fungi. Trends in Plant Science 18:484-491.

Cheplick, G. P. 2005. Biomass partitioning and reproductive allocation in the invasive, cleistogamous grass *Microstegium vimineum*: Influence of the light environment. Journal of the Torrey Botanical Society 132:214-224.

Cheplick, G. P. 2008. Growth trajectories and size-dependent reproduction in the highly invasive grass *Microstegium vimineum*. Biological Invasions 10:761-770.

Cheplick, G. P. 2010. Limits to local spatial spread in a highly invasive annual grass (*Microstegium vimineum*). Biological Invasions 12:1759-1771.

Cheplick, G. P. 2015. Population differentiation in the tiller architecture of *Microstegium vimineum* (Poaceae) in relation to habitat. Plant Species Biology 30:16-27.

Cheplick, G. P., and J. Fox. 2011. Density-dependent growth and reproduction of *Microstegium vimineum* in contrasting light environments. Journal of the Torrey Botanical Society 138:62-72.

Christen, D. C., and G. R. Matlack. 2009. The habitat and conduit functions of roads in the spread of three invasive plant species. Biological Invasions 11:453-465.

Cipollini, K., and M. G. Bohrer. 2016. Comparison of allelopathic effects of five invasive species on two native species. Journal of the Torrey Botanical Society 143:427-436.

Claridge, K., and S. B. Franklin. 2002. Compensation and plasticity in an invasive plant species. Biological Invasions 4:339-347.

Cole, P. G., and J. F. Weltzin. 2004. Environmental correlates of the distribution and abundance of *Microstegium vimineum*, in east Tennessee. Southeastern Naturalist 3:545-562.

Corbett, B. F., and J. A. Morrison. 2012. The allelopathic potentials of the non-native invasive plant *Microstegium vimineum* and the native *Agerantina altissima*: two dominant species of the eastern forest herb layer. Northeastern Naturalist 19:297-312.

Craig, M. E., and J. M. Fraterrigo. 2017. Plant-microbial competition for nitrogen increases microbial activities and carbon loss in invaded soils. Oecologia 184:583-596.

Craig, M. E., N. Lovko, S. L. Flory, J. P. Wright, and R. P. Phillips. 2019. Impacts of an invasive grass on soil organic matter pools vary across a tree-mycorrhizal gradient. Biogeochemistry 144:149-164.

Culpepper, L. Z., H. H. Wang, T. E. Koralewski, W. E. Grant, and W. E. Rogers. 2018. Understory upheaval: factors influencing Japanese stiltgrass invasion in forestlands of Tennessee, United States. Botanical Studies 59:20.

Dabney, S. M., J. D. Schreiber, C. S. Rothrock, and J. R. Johnson. 1996. Cover crops affect sorghum seedling growth. Agronomy Journal 88:961-970.

Daehler, C. C. 2003. Performance comparisons of co-occurring native and alien invasive plants: implications for conservation and restoration. Annual Review of Ecology Evolution and Systematics 34:183-211.

Daughtridge, A. T., S. R. Boese, S. G. Pallardy, and H. A. Garrett. 1986. A rapid staining technique for assessment of ectomycorrhizal infection of oak roots. Canadian Journal of Botany 64:1101-1103.

DeMeester, J. E., and D. D. Richter. 2010. Differences in wetland nitrogen cycling between the invasive grass *Microstegium vimineum* and a diverse plant community. Ecological Applications 20:609-619.

Dickie, I. A., R. T. Koide, and K. C. Steiner. 2002. Influences of established trees on mycorrhizas, nutrition, and growth of *Quercus rubra* seedlings. Ecological Monographs 72:505-521.

Dormann, C. F., J. M. Calabrese, G. Guillera-Arroita, E. Matechou, V. Bahn, K. Bartoń, C. M. Beale, S. Ciuti, J. Elith, K. Gerstner, J. Guelat, P. Keil, J. J. Lahoz-Monfort, L. J. Pollock, B. Reineking, D. R. Roberts, B. Schröder, W. Thuiller, D. I. Warton, B. A. Wintle, S. N. Wood, R. O. Wüest, and F. Hartig. 2018. Model averaging in ecology: A review of Bayesian, information-theoretic, and tactical approaches for predictive inference. Ecological Monographs, 88:485-504.

Droste, T., S. L. Flory, and K. Clay. 2010. Variation for phenotypic plasticity among populations of an invasive exotic grass. Plant Ecology 207:297-306.

Duchesneau, K., A. Golemiee, R. L. Colautti, and P. M. Antunes. 2021. Functional shifts of soil microbial communities associated with *Alliaria petiolate* invasion. Pedobiologia 84:50700.

Durner, G. M., and J. E. Gates. 1993. Spatial ecology of black rat snakes on Remington Farms, Maryland. Journal of Wildlife Management 57:812-826.

Ehrenfeld, J. G., P. Kourtev, and W. Z. Huang. 2001. Changes in soil functions following invasions of exotic understory plants in deciduous forests. Ecological Applications 11:1287-1300.

Ehrenfeld, J. G., and N. Scott. 2001. Invasive species and the soil: effects on organisms and ecosystems processes. Ecological Applications 11:1259-1260.

Elgersma, K. J., S. Yu, V. Torsten, and J. G. Ehrenfeld. 2012. Microbial-mediated feedbacks of leaf litter on invasive plant growth and interspecific competition. Plant and Soil 356:341-355.

Ellenberg, H. 1988: Vegetation ecology of central Europe. Cambridge: Cambridge University Press.

Emery, S. M., and J. A. Rudgers. 2014. Biotic and abiotic predictors of ecosystem engineering traits of the dune building grass, *Ammophila breviligulata*. Ecosphere 5.

Emery, S. M., J. Uwimbabazi, and S. L. Flory. 2011. Fire intensity effects on seed germination of native and invasive Eastern deciduous forest understory plants. Forest Ecology and Management **261**:1401-1408.

Engel, M., A. Behnke, S. Bauerfeld, C. Bauer, C. Buschbaum, N. Volkenborn, and T. Stoeck. 2012. Sample pooling sbscures diversity patterns in intertidal ciliate community composition and structure. FEMS Microbial Ecology 79:741-750.

Enders, M., M.-T. Huett, and J. M. Jeschke. 2018. Drawing a map of invasion biology based on a network of hypotheses. Ecosphere 9.

Fairbrothers, D. E., and J. R. Gray. 1972. *Microstegium vimineum* (Trin.) A. Camus (Gramineae) in United States. Bulletin of the Torrey Botanical Club 99:97-+.

Flory, S. L. 2010. Management of *Microstegium vimineum* invasions and recovery of resident plant communities. Restoration Ecology 18:103-112.

Flory, S. L., J. Bauer, R. P. Phillips, and K. Clay. 2017. Effects of a non-native grass invasion decline over time. Journal of Ecology 105:1475-1484.

Flory, S. L., and K. Clay. 2009. Invasive plant removal method determines native plant community responses. Journal of Applied Ecology 46:434-442.

Flory, S. L., and K. Clay. 2010a. Non-native grass invasion alters native plant composition in experimental communities. Biological Invasions 12:1285-1294.

Flory, S. L., and K. Clay. 2010b. Non-native grass invasion suppresses forest succession. Oecologia 164:1029-1038.

Flory, S. L., and K. Clay. 2013. Pathogen accumulation and long-term dynamics of plant invasions. Journal of Ecology 101:607-613.

Flory, S. L., K. Clay, S. M. Emery, J. R. Robb, and B. Winters. 2015. Fire and non-native grass invasion interact to suppress tree regeneration in temperate deciduous forests. Journal of Applied Ecology 52:992-1000.

Flory, S. L., F. R. Long, and K. Clay. 2011. Greater performance of introduced vs. native range populations of *Microstegium vimineum* across different light environments Basic and Applied Ecology 12:350-359.

Fraterrigo, J. M., M. S. Strickland, A. D. Keiser, and M. A. Bradford. 2011. Nitrogen uptake and preference in a forest understory following invasion by an exotic grass. Oecologia 167:781-791.

Gallery, R.E., J. W. Dalling, and A. E. Arnold. 2007. Diversity, host affinity and distribution of seed-infecting fungi: a case-study with neotropical Cecropia. Ecology, 83:582–588.

Giba, Z., D. Grubisic, and R. Konjevic. 2003. Nitrogen oxides as environmental sensors for seeds. Seed Science Research 13:187-196.

Gibson, D. J., G. Spyreas, and J. Benedict. 2002. Life history of *Microstegium vimineum* (Poaceae), an invasive grass in southern Illinois. Journal of the Torrey Botanical Society 129:207-219.

Hartman, K. M., and B. C. McCarthy. 2008. Changes in forest structure and species composition following invasion by a non-indigenous shrub, Amur honeysuckle (Lonicera maackii). Journal of the Torrey Botanical Society 135:245-259.

Hartnett, D. C., and G. W. T. Wilson. 1999. Mycorrhizae influence plant community structure and diversity in tallgrass prairie. Ecology 80:1187-1195.

Hengl, T., and H. I. Reuter 2009. Geomorphometry: concepts, software, applications. Elsevier, Amsterdam, The Netherlands.

Holopainen, T., and S. Vaittinen. 1988. Observation on Scots pine mycorrhizae in the surroundings of a fluting mill. Karstenia 28:35-39.

Horton, J. L., and H. S. Neufeld. 1998. Photosynthetic responses of *Microstegium vimineum* (Trin.) A. Camus, a shade-tolerant, C-4 grass, to variable light environments. Oecologia 114:11-19.

Howe, H. F., J. S. Brown, and B. Zorn-Arnold. 2002. A rodent plague on prairie diversity. Ecology Letters 5:30-36.

Huebner, C. D. 2010. Spread of an invasive grass in closed-canopy deciduous forests across local and regional environmental gradients. Biological Invasions 12:2081-2089.

Huebner, C. D. 2011. Seed mass, viability, and germination of Japanese stiltgrass (*Microstegium vimineum*) under variable light and moisture conditions. Invasive Plant Science and Management 4:274-283.

Hurlbert, S. H. 1984. Pseduoreplication and the design of ecological field experiments. Ecological Monographs 54:187-211.

Jeschke, J. M. 2014. General hypotheses in invasion ecology. Diversity and Distributions 20:1229-1234.

Johnson, D. J., S. L. Flory, A. Shelton, C. Huebner, and K. Clay. 2015. Interactive effects of a non-native invasive grass *Microstegium vimineum* and herbivore exclusion on experimental tree regeneration under differing forest management. Journal of Applied Ecology 52:210-219.

Judge, C. A., J. C. Neal, and T. H. Shear. 2008. Japanese stiltgrass (*Microstegium vimineum*) management for restoration of native plant communities. Invasive Plant Science and Management 1:111-119.

Kendig, A. E., V. J. Svahnström, A. Adhikari, P. F. Harmon, and S. L. Flory. 2021. Emerging fungal pathogen of an invasive grass: implications for competition with native plant species. Plos One 16:e0237894.

Kerns, B. K., C. Tortorelli, M. A. Day, T. Nietupski, A. M. G. Barros, J. B. Kim, and M. A. Krawchuk. 2020. Invasive grasses: A new perfect storm for forested ecosystems? Forest Ecology and Management 463.

Kleczewski, N. M., S. L. Flory, and K. Clay. 2012. Variation in pathogenicity and host range of *Bipolaris* sp. causing leaf blight disease on the invasive grass Microstegium vimineum. Weed Science 60:486-493.

Klironomos, J. N. 2002. Feedback with soil biota contributes to plant rarity and invasiveness in communities. Nature 417:67-70.

Koenig, K., and S. M. Emery 2021. Abiotic factors, not mycorrhizal associations, predict size and abundance of the invasive grass *Microstegium vimineum*. Journal of the Torrey Botanical Society 148:294-307.

Kourtev, P. S., J. G. Ehrenfeld, and M. Haggblom. 2002. Exotic plant species alter the microbial community structure and function in the soil. Ecology 83:3152-3166.

Kourtev, P. S., J. G. Ehrenfeld, and W. Z. Huang. 1998. Effects of exotic plant species on soil properties in hardwood forests of New Jersey. Water Air and Soil Pollution 105:493-501.

Kramer, T. D., R. J. Warren, Y. Y. Tang, and M. A. Bradford. 2012. Grass invasions across a regional gradient are associated with declines in belowground carbon pools. Ecosystems 15:1271-1282.

Kumar, A., R. P. Phillips, A. Scheibe, S. Klink, and J. Pausch. 2020. Organic matter priming by invasive plants depends on dominant mycorrhizal association. Soil Biology and Biochemistry 40:07645.

Kuznetsova, A., P. B. Brockhoff, and R. H. B. Christensen. 2017. lmerTest Package: Tests in Linear Mixed Effects Models. Journal of Statistical Software 82:1-26.

Lau, J. A., and E. H. Schultheis. 2015. When two invasion hypotheses are better than one. New Phytologist 205:958-960.

Le, T. S., P. T. K. Thoa, and N. V. Tuan. 2019. GIS-based habitat model to predict potential areas for the upcoming occurrences of an alien invasive plant, *Mimosa pigra* L. Forestry Studies 70:31-43.

Lee, M. R., S. L. Flory, and R. P. Phillips. 2012. Positive feedbacks to growth of a invasive grass through alteration of nitrogen cycling. Oecologia 170:457-465.

Lee, M. R., C. Tu, X. Chen, and S. J. Hu. 2014. Arbuscular mycorrhizal fungi enhance P uptake and alter plant morphology in the invasive plant *Microstegium vimineum.* Biological Invasions 16:1083-1093.

Leicht, S. A., J. A. Silander, and K. Greenwood. 2005. Assessing the competitive ability of Japanese stilt grass, *Microstegium vimineum* (Trin.) A. Camus. Journal of the Torrey Botanical Society 132:573-580.

Lekberg, Y., J. D. Bever, R. A. Bunn, R. M. Callaway, M. M. Hart, S. N. Kivlin, J. Klironomos, B. G. Larkin, J. L. Maron, K. O. Reinhart, M. Remke, and W. H. van der Putten. 2018. Relative importance of competition and plant-soil feedback, their synergy, context dependency and implications for coexistence. Ecology Letters 21:1268-1281.

Lemke, D., P. E. Hulme, J. A. Brown, and W. Tadesse. 2011. Distribution modelling of Japanese honeysuckle (*Lonicera japonica*) invasion in the Cumberland Plateau and Mountain Region, USA. Forest Ecology and Management 262:139-149.

Length, R. V. 2022. Emmeans: Estimated marginal means, aka least-squares means. R package version 1.8.1-1.

Levine, J. M., M. Vila, C. M. D'Antonio, J. S. Dukes, K. Grigulis, and S. Lavorel. 2003. Mechanisms underlying the impacts of exotic plant invasions. Proceedings of the Royal Society B-Biological Sciences 270:775-781.

Li, X., D. Li, Y. Li, Z. Ma, and T. Zhai. 2002. Habitat evaluation for crested ibis: A GIS-based approach. Ecological Research 17:565-573.

Li, X., M. Song, X. Yao, Q. Chai, W. R. Simpson, C. Li, and Z. Nan. 2017. The effect of seed-borne fungi and Epichloë endophyte on seed germination and biomass of *Elymus sibiricus*. Frontiers in Microbiology 8:e2488.

Liebhold, A., E. G. Brockerhoff, S. Kalisz, M. A. Nunez, D. A. Wardle, and M. J. Wingfield. 2017. Biological invasions in forest ecosystems. Biological Invasions 19:3437-3458.

Lou, Y., A. S. Davis, and A. C. Yannarell. 2016. Interactions between allelochemicals and the microbial community affect weed suppression following cover crop residue incorporation into soil. Plant Soil 399:357-371.

Luna, B., and J. M. Moreno. 2009. Light and nitrate effects on seed germination of Mediterranean plant species of several functional groups. Plant Ecology 2003:123-135.

Mack, K. M. L., and J. D. Bever. 2014. Coexistence and relative abundance in plant communities are determined by feedbacks when the scale of feedback and dispersal is local. Journal of Ecology 102:1195-1201.

Mack, R. N., D. Simberloff, W. M. Lonsdale, H. Evans, M. Clout, and F. A. Bazzaz. 2000. Biotic invasions: causes, epidemiology, global consequences, and control. Ecological Applications 10:689-710.

Maringer, A., and L. Slotta-Bachmayr. 2006. A GIS-based habitat-suitability model as a tool for the management of beavers *Castor fiber*. *Acta Theriologica* 51:373-382.

Marshall, J. M., and D. S. Buckley. 2008. Influence of litter removal and mineral soil disturbance on the spread of an invasive grass in a Central Hardwood forest. Biological Invasions 10:531-538.

Martin, P. H., C. D. Canham, and P. L. Marks. 2009. Why forests appear resistant to exotic plant invasions: intentional introductions, stand dynamics, and the role of shade tolerance. Frontiers in Ecology and the Environment 7:142-149.

McCollin, D. 1998. Forest edges and habitat selection in birds: a functional approach. Ecography 21:247-260.

McGonigle, T. P., M. H. Miller, D. G. Evans, G. L. Fairchild, and J. A. Swan. 1990. A new method which gives an objective measure of colonization of roots by vesicular arbuscular mycorrhizal fungi. New Phytologist 115:495-501.

McGrath, D. A., and M. A. Binkley. 2009. *Microstegium vimineum* invasion changes soil chemistry and microarthropod communities in Cumberland Plateau Forests. Southeastern Naturalist 8:141-156.

Medley, K. E. 1997. Distribution of the non-native shrub Lonicera maackii in Kramer Woods, Ohio. Physical Geography 18:18-36.

Miller, N. P., and G. R. Matlack. 2010. Population expansion in an invasive grass, *Microstegium vimineum*: a test of the channeled diffusion model. Diversity and Distributions 16:816-826.

Moeslund, J. E., L. Arge, P. K. Bocher, T. Dalgaard, M. V. Odgaard, B. Nygaard, and J. Svenning. 2013. Topographically controlled soil moisture is the primary driver of local vegetation patterns across a lowland region. Ecosphere 4:1-26.

Muff, S., E. B. Nilsen, R. B. O’Hara, and C R. Nater. 2022. Rewriting results sections in the language of evidence. Trends in Ecology & Evolution 37:203-210.

Nord, A. N., D. A. Mortensen, and E. S. J. Rauschert. 2010. Environmental factors influence early population growth of Japanese stiltgrass. Invasive Plant Science and Management 3:17-25.

North, B. A., and A. P. Torzilli. 2017. Characterization of the root and soil microbiome associated with invasive Microstegium vimineum in the presence and absence of a native plant community. Botany 95:513-520.

Novy, A., S. L. Flory, and J. M. Hartman. 2013. Evidence for rapid evolution of phenology in an invasive grass. Journal of Evolutionary Biology 26:443-450.

Oswalt, C. M., S. N. Oswalt, and W. K. Clatterbuck. 2007. Effects of *Microstegium vimineum* (Trin.) A. Camus on native woody species density and diversity in a productive mixed-hardwood forest in Tennessee. Forest Ecology and Management 242:727-732.

Pérez-Fernández, M. A., E. Calvo-Magro, J. Montanero-Fernández, and J. A. Oyola-Velasco. 2006. Seed germination in response to chemicals: effect of nitrogen and pH in the media. Journal of Environmental Biology 27:13-20.

Pisula, N. L., and S. J. Meiners. 2010. Relative allelopathic potential of invasive plant species in a young disturbed woodland. Journal of the Torrey Botanical Society 137:81-87.

Price, G. J., and A. C. Blair. 2019. Garlic mustard as a potential driver of change in soil microbial community metabolism under controlled conditions. Bios 90:218-226.

Rauschert, E. S. J., D. A. Mortensen, O. N. Bjornstad, A. N. Nord, and N. Peskin. 2010. Slow spread of the aggressive invader, *Microstegium vimineum* (Japanese stiltgrass). Biological Invasions 12:563-579.

Redman, D. E. 1995. Distribution and habitat types for Nepal [*Microstegium vimineum* (Trin.) Camus] in Maryland and the District of Columbia. Castanea 60:270-275.

Reinhart, K. O., and R. M. Callaway. 2006. Soil biota and invasive plants. New Phytologist 170:445-457.

Reinhart, K. O., A. Packer, W. H. van der Putten, and K. Clay. 2003. Plant-soil biota interactions and spatial distribution of black cherry in its native and invasive ranges. Ecology Letters 6:1046-1050.

Rinella, M. J., and K. O. Reinhart. 2018. Toward more robust plant-soil feedback research. Ecology 99:550-556.

Rippel, T. M., C. L. Iosue, P. J. Succi, D. D. Wykoff, and S. K. Chapman. 2020. Comparing the impacts of an invasive grass on nitrogen cycling and ammonia-oxidizing prokaryotes in high-nitrogen forests, open fields, and wetlands. Plant and Soil 449:65-71.

Ross, K. A., J. G. Ehrenfeld, and M. V. Patel. 2011. The effects of nitrogen addition on the growth of two exotic and two native forest understory plants. Biological Invasions 13:2203-2216.

Ruckli, R., H. P. Rusterholz, and B. Baur. 2013. Invasion of Impatiens glandulifera affects terrestrial gastropods by altering microclimate. Acta Oecologica-International Journal of Ecology 47:16-23.

Sage, R. F., and S. Sultmanis. 2016. Why are there no C-4 forests? Journal of Plant Physiology 203:55-68.

Saumel, I., and I. Kowarik. 2010. Urban rivers as dispersal corridors for primarily wind-dispersed invasive tree species. Landscape and Urban Planning 94:244-249.

Schramm, J. W., and J. G. Ehrenfeld. 2012. Patterns of patch colonization and expansion in the non-native grass *Microstegium vimineum* (Poaceae). Rhodora 114:1-20.

Setterfield, S. A., P. J. Clifton, L. B. Hutley, N. A. Rossiter-Rachor, and M. M. Douglas. 2018. Exotic grass invasion alters microsite conditions limiting woody recruitment potential in an Australian savanna. Scientific Reports 8.

Shannon, S., S. L. Flory, and H. Reynolds. 2012. Competitive context alters plant-soil feedback in an experimental woodland community. Oecologia 169:235-243.

Shannon-Firestone, S., H. L. Reynolds, R. P. Phillips, S. L. Flory, and A. Yannarell. 2015. The role of ammonium oxidizing communities in mediating effects of an invasive plant on soil nitrification. Soil Biology & Biochemistry 90:266-274.

Shoemaker, C. A., and W. H. Carlson. 1990. pH affects seed germination of eight bedding plant species. Horticultural Science 25:762-764.

Smith, L. M. 2013. Extended leaf phenology in deciduous forest invaders: mechanisms of impact on native communities. Journal of Vegetation Science 24:979-987.

Suding, K. N., W. S. Harpole, T. Fukami, A. Kulmatiski, A. S. MacDougall, S. Stein, W. H. van der Putten. 2013. Consequences of plant-soil feedbacks in invasion. Journal of Ecology 101:298-308.

Strickland, M. S., J. L. DeVore, J. C. Maerz, and M. A. Bradford. 2011. Loss of faster-cycling soil carbon pools following grass invasion across multiple forest sites. Soil Biology & Biochemistry 43:452-454.

Tekiela, D. R., and J. N. Barney. 2013. Quantifying *Microstegium vimineum* seed movement by non-riparian water dispersal using an ultraviolet-marking based recapture method. *Plos One* 8.

Thompson, R., and B. M. Starzomski. 2007. What does biodiversity actually do? A review for managers and policy makers. Biodiversity and Conservation 16:1359-1378.

Tickner, D. P., P. G. Angold, A. M. Gurnell, and J. O. Mountford. 2001. Riparian plant invasions: hydrogeomorphological control and ecological impacts. Progress in Physical Geography 25:22-52.

Touchette, B. W., and G. A. Romanello. 2010. Growth and water relations in a central North Carolina population of *Microstegium vimineum* (Trin.) A. Camus. Biological Invasions 12:893-903.

U. S. Fish and Wildlife. 1981. Ecological Services Manual: Development of Habitat Suitability Index Models. Retrieved from https://www.fws.gov/policy/esm103.pdf.

U. S. Geological Survey. 2022. National Water Information System for McAlpine Dam at Louisville, KY. Retrieved from http://waterdata.usgs.org on September 21, 2022.

van der Putten, W. H. 2002. How to be invasive. Nature 417:32– 33.

van der Putten, W. H., R. D. Bardgett, J. D. Bever, T. M. Bezemer, J. A. Schweitzer, K. N. Suding, T. F. J. Van de Voorde, and D. A. Wardle. 2013. Plant-soil feedbacks: the past, the present and future challenges. Journal of Ecology 101:265-276.

Vasquez, E., R. Sheley, and T. Svejcar. 2008. Creating invasion resistant soils via nitrogen management. Invasive Plant Science and Management 1:304-314.

Vogelsang, K. M., and J. D. Bever. 2009. Mycorrhizal densities decline in association with nonnative plants and contribute to plant invasion. Ecology 90:399-407.

Vogelsang, K. M., H. L. Reynolds, and J. D. Bever. 2006. Mycorrhizal fungal identity and richness determine the diversity and productivity of a tallgrass prairie system. New Phytologist 172:554-562.

Vogiatzakis, I. N., and G. H. Griffiths. 2006. A GIS-based empirical model for vegetation prediction in Lefka Ori, Crete. Plant Ecology 183:311-323.

Ward, J. S., and T. L. Mervosh. 2012. Nonchemical and herbicide treatments for management of Japanese stiltgrass (*Microstegium vimineum*). Invasive Plant Science and Management 5:9-19.

Wardle, D. A., R. D. Bardgett, R. M. Callaway, and W. H. Van der Putten. 2011. Terrestrial Ecosystem Responses to Species Gains and Losses. Science 332:1273-1277.

Warren, R. J., V. Bahn, and M. A. Bradford. 2012. The interaction between propagule pressure, habitat suitability and density-dependent reproduction in species invasion. Oikos 121:874-881.

Warren, R. J., V. Bahn, T. D. Kramer, Y. Y. Tang, and M. A. Bradford. 2011a. Performance and reproduction of an exotic invader across temperate forest gradients. Ecosphere 2(2):art14.

Warren, R. J., J. P. Wright, and M. A. Bradford. 2011b. The putative niche requirement and landscape dynamics of *Microstegium vimineum*: an invasive Asian grass. Biological Invasions 13:471-483.

Watling, J. I., C. R. Hickman, and J. L. Orrock. 2011. Invasive shrub alters native forest amphibian communities. Biological Conservation 144:2597-2601.

Webster, C. R., J. H. Rock, R. E. Froese, and M. A. Jenkins. 2008. Drought-herbivory interaction disrupts competitive displacement of native plants by *Microstegium vimineum*, 10-year results. Oecologia 157:497-508.

Wilson, C. H., T. T. Caughlin, D. J. Civitello, and S. L. Flory. 2015. Combining mesocosm and field experiments to predict invasive plant performance: a hierarchical Bayesian approach. Ecology 96:1084-1092.

Winter, K., M. R. Schmitt, and G. E. Edwards. 1982. *Microstegium vimineum*, a shade adapted C4 grass. Plant Science Letters 24:311-318.

Wolfe, B. E., and J. N. Klironomos. 2005. Breaking new ground: soil communities and exotic slant invasion. Bioscience 55:477-487.

Woods, A. J., Omernik J. M., Brockman C. S., Gerber T. D., Hosteter W. D., and Azevedo S. H. 2002-a Ecoregions of Indiana and Ohio (color poster with map, descriptive text, summary tables, and photographs): Reston, VA., U.S. Geological Survey (map scale 1:1,000,000). Retrieved from https://store.usgs.gov/assets/MOD/StoreFiles/Ecoregion/21631_in_oh_front.pdf.

Woods, A. J., Omernik J. M., Martin W. H., Pond G. J., Andrews W. M., Call S. M., Comstock J. A., and Taylor D. D. 2002-b. Ecoregions of Kentucky (color poster with map, descriptive text, summary tables, and photographs): Reston, VA., U.S. Geological Survey (map scale 1:1,000,000). Retrieved from https://store.usgs.gov/assets/MOD/StoreFiles/Ecoregion/113780_ky_front.pdf.

Yamazaki, M., S. Iwamoto, and K. Seiwa. 2009. Distance- and density-dependent seedling mortality caused by several diseases in eight tree species co-occurring in a temperate forest. Plant Ecology 201:181-196.

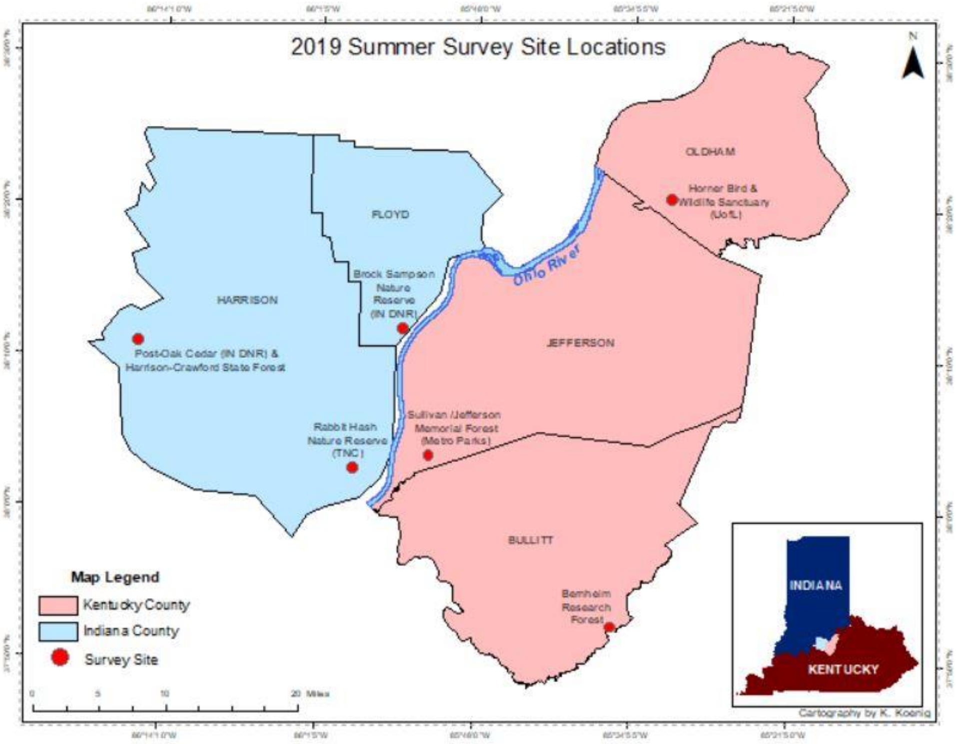
Zhong, M., Y. Miao, S. Han, and D. Wang. 2019. Nitrogen addition decreases seed germination in a temperate steppe. Ecology and Evolution 9(15):8441-8449.

Ziska, L. H., M. B. Tomecek, M. Valerio, and J. P. Thompson. 2015. Evidence for recent evolution in an invasive species, *Microstegium vimineum*, Japanese stiltgrass. Weed Research 55:260-267.

APPENDIX I

SUPPLEMENTAL DATA FROM CHAPTER II

Appendix I Figure 16: Map of survey site locations.



Appendix I Table 7: summary of survey site locations.

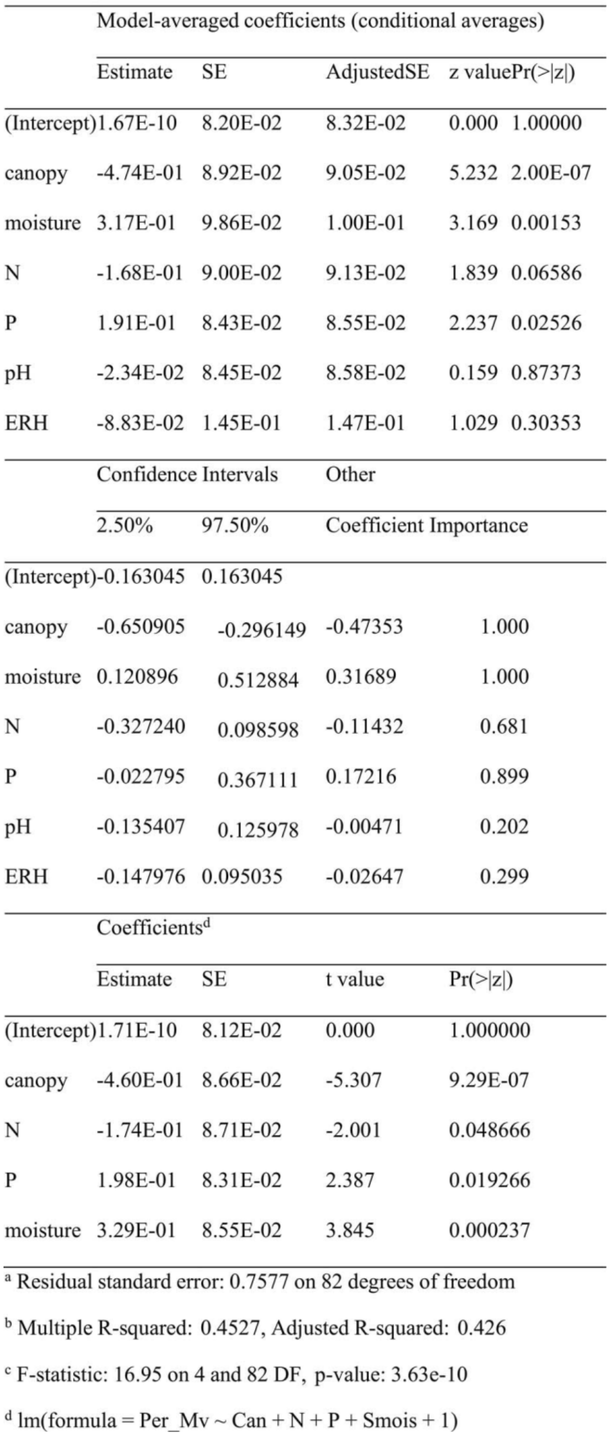
Property Name	**Owner/ Manager**	**Latitude/ Longitude**	**Size of Property**	**Extent of Invasion**	**Managed for Invasive Plants**	**Managed for *Microstegium***
Horner Bird & Wildlife Sanctuary	University of Louisville	38.3445135 -085.5309699	85 ha	Major	No	No
Bernheim Research Forest	Bernheim Arboretum	37.8718134 -085.6101163	6530 ha	Medium	Yes	Yes mowing
Jefferson Memorial Forest – Sullivan Property	Louisville Metro Parks	38.0635972 -085.8665012	2630 ha	Major	Yes	No
Rabbit Hash Glade Nature Reserve	The Nature Conservancy	38.0458196 -085.9663535	66 ha	Minor	Yes	Yes herbicide
Brock Sampson Nature Reserve	Indiana DNR	38.2003067 -085.9053065	247 ha	Major	Yes	No
Post Oak Cedar Reserve / Harrison-Crawford State Forest	Indiana DNR	38.1830374 -085.2781358	91 ha	Minor	Yes	Yes, herbicide and mowing

Appendix I Table 8: Variable ranges for each location and site.

Variable	Horner Bird and Wildlife Sanctuary	Bernheim Research Forest	Jefferson Memorial Forest, Sullivan Property	Rabbit Hash Glade Nature Reserve	Brock Sampson Nature Reserve	Post Oak Cedar/ Harrison-Crawford State Forest
Canopy cover (%)	63.08–98.70	82.10–98.20	0.16–96.10	91.42–97.14	75.04–98.44	83.10–98.44
Soil moisture (%)	7.6–19.1	18.7–24.6	8.3–24.2	2.6–12.7	0.8–16.9	4.9–17.5
Soil nitrogen (%)	0.109–0.540	0.140–0.366	0.098–0.290	0.119–0.354	0.129–0.460	0.149–0.487
Soil phosphorous (lb/acre)	5–36	5–15	10–54	3–19	8–109	5–23
Soil pH	5.0–7.8	7.1–7.7	5.2–7.9	4.5–6.7	4.5–7.0	5.5–7.8
Soil extraradical hyphal abundance (cm g^{-1})	233–594	247–564	112–536	213–599	110–918	224–765
Root colonization (%)	40–90	60–100	5–95	20–60	15–85	30
Microstegium vimineum tiller height (cm)	8.73–54.29	9.05–48.45	8.13–59.37	33.40–54.17	19.37–53.98	88.5–93.6
Microstegium vimineum cover (%)	0–95	0–70	0–95	5–10	0–85	5–10
No. of plots	16	8	15	16	16	16

Appendix I Table 9: Model-average coefficients and formula values.

Table 9A: Percent Cover



	Model-averaged coefficients (conditional averages)				
	Estimate	SE	AdjustedSE	z value	Pr(>|z|)
(Intercept)	1.67E-10	8.20E-02	8.32E-02	0.000	1.00000
canopy	-4.74E-01	8.92E-02	9.05E-02	5.232	2.00E-07
moisture	3.17E-01	9.86E-02	1.00E-01	3.169	0.00153
N	-1.68E-01	9.00E-02	9.13E-02	1.839	0.06586
P	1.91E-01	8.43E-02	8.55E-02	2.237	0.02526
pH	-2.34E-02	8.45E-02	8.58E-02	0.159	0.87373
ERH	-8.83E-02	1.45E-01	1.47E-01	1.029	0.30353

	Confidence Intervals		Other	
	2.50%	97.50%	Coefficient	Importance
(Intercept)	-0.163045	0.163045		
canopy	-0.650905	-0.296149	-0.47353	1.000
moisture	0.120896	0.512884	0.31689	1.000
N	-0.327240	0.098598	-0.11432	0.681
P	-0.022795	0.367111	0.17216	0.899
pH	-0.135407	0.125978	-0.00471	0.202
ERH	-0.147976	0.095035	-0.02647	0.299

	Coefficients[d]			
	Estimate	SE	t value	Pr(>|z|)
(Intercept)	1.71E-10	8.12E-02	0.000	1.000000
canopy	-4.60E-01	8.66E-02	-5.307	9.29E-07
N	-1.74E-01	8.71E-02	-2.001	0.048666
P	1.98E-01	8.31E-02	2.387	0.019266
moisture	3.29E-01	8.55E-02	3.845	0.000237

[a] Residual standard error: 0.7577 on 82 degrees of freedom

[b] Multiple R-squared: 0.4527, Adjusted R-squared: 0.426

[c] F-statistic: 16.95 on 4 and 82 DF, p-value: 3.63e-10

[d] lm(formula = Per_Mv ~ Can + N + P + Smois + 1)

Table 9B: Tiller Height

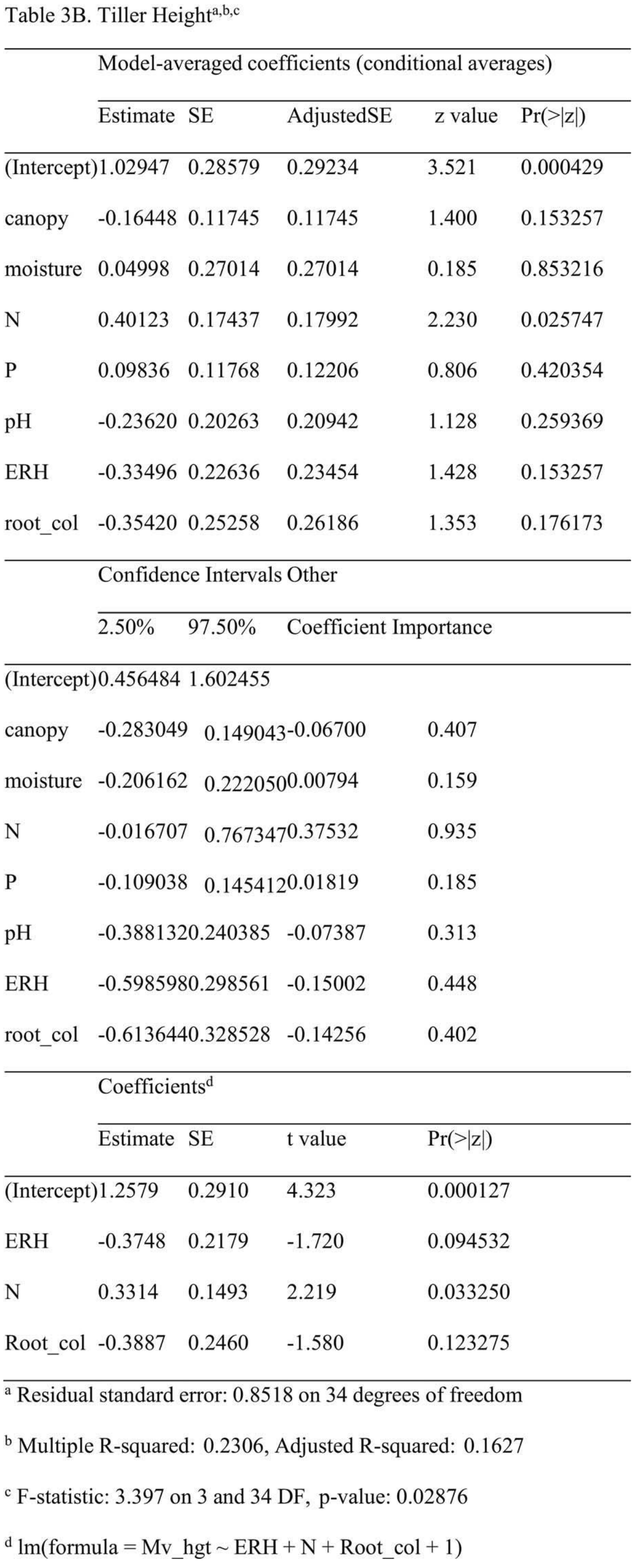


Table 3B. Tiller Height[a,b,c]

	Model-averaged coefficients (conditional averages)				
	Estimate	SE	AdjustedSE	z value	Pr(>|z|)
(Intercept)	1.02947	0.28579	0.29234	3.521	0.000429
canopy	-0.16448	0.11745	0.11745	1.400	0.153257
moisture	0.04998	0.27014	0.27014	0.185	0.853216
N	0.40123	0.17437	0.17992	2.230	0.025747
P	0.09836	0.11768	0.12206	0.806	0.420354
pH	-0.23620	0.20263	0.20942	1.128	0.259369
ERH	-0.33496	0.22636	0.23454	1.428	0.153257
root_col	-0.35420	0.25258	0.26186	1.353	0.176173

	Confidence Intervals		Other	
	2.50%	97.50%	Coefficient	Importance
(Intercept)	0.456484	1.602455		
canopy	-0.283049	0.149043	-0.06700	0.407
moisture	-0.206162	0.222050	0.00794	0.159
N	-0.016707	0.767347	0.37532	0.935
P	-0.109038	0.145412	0.01819	0.185
pH	-0.388132	0.240385	-0.07387	0.313
ERH	-0.598598	0.298561	-0.15002	0.448
root_col	-0.613644	0.328528	-0.14256	0.402

	Coefficients[d]			
	Estimate	SE	t value	Pr(>|z|)
(Intercept)	1.2579	0.2910	4.323	0.000127
ERH	-0.3748	0.2179	-1.720	0.094532
N	0.3314	0.1493	2.219	0.033250
Root_col	-0.3887	0.2460	-1.580	0.123275

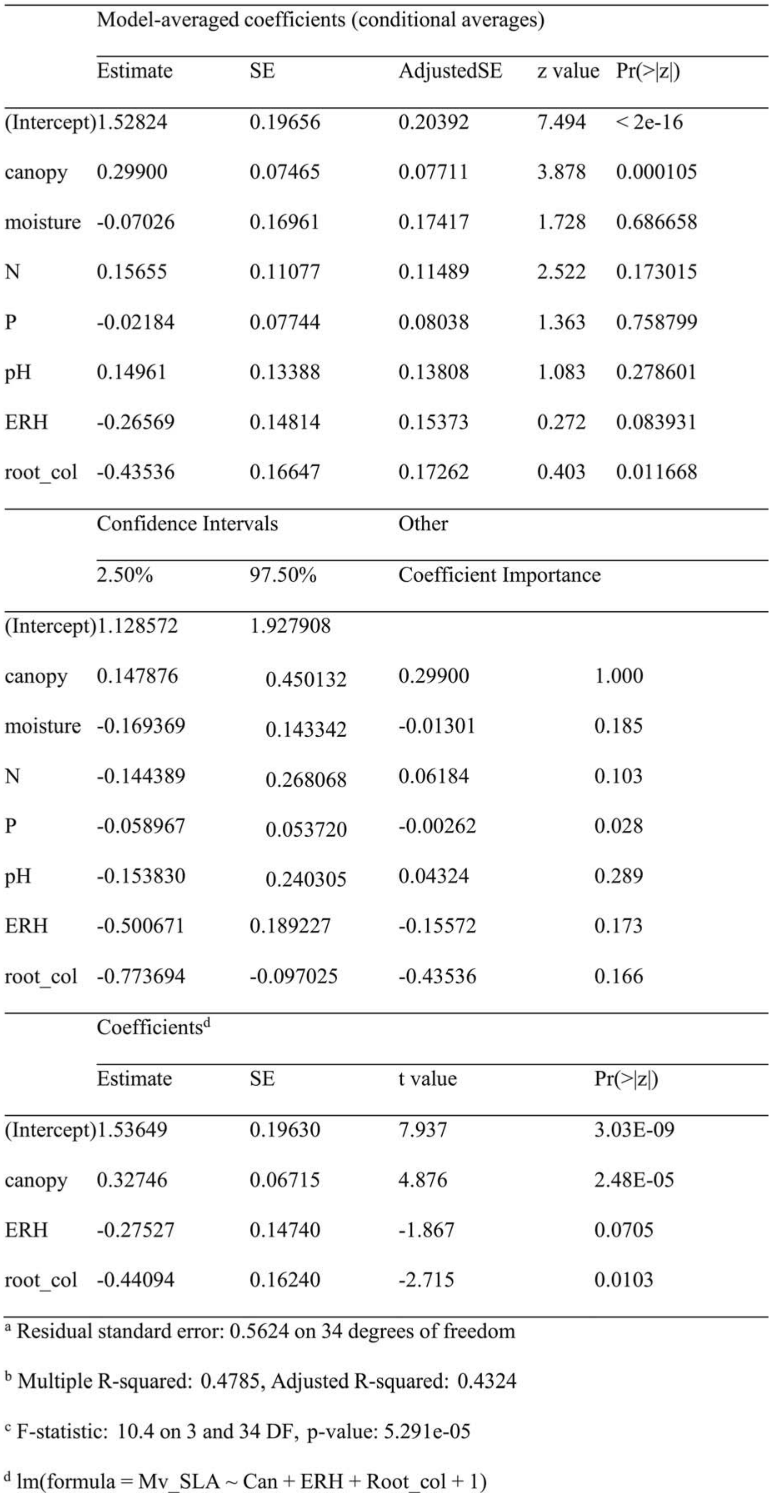
[a] Residual standard error: 0.8518 on 34 degrees of freedom

[b] Multiple R-squared: 0.2306, Adjusted R-squared: 0.1627

[c] F-statistic: 3.397 on 3 and 34 DF, p-value: 0.02876

[d] lm(formula = Mv_hgt ~ ERH + N + Root_col + 1)

Table 9C: SLA



	Model-averaged coefficients (conditional averages)				
	Estimate	SE	AdjustedSE	z value	Pr(>|z|)
(Intercept)	1.52824	0.19656	0.20392	7.494	< 2e-16
canopy	0.29900	0.07465	0.07711	3.878	0.000105
moisture	-0.07026	0.16961	0.17417	1.728	0.686658
N	0.15655	0.11077	0.11489	2.522	0.173015
P	-0.02184	0.07744	0.08038	1.363	0.758799
pH	0.14961	0.13388	0.13808	1.083	0.278601
ERH	-0.26569	0.14814	0.15373	0.272	0.083931
root_col	-0.43536	0.16647	0.17262	0.403	0.011668

	Confidence Intervals		Other	
	2.50%	97.50%	Coefficient Importance	
(Intercept)	1.128572	1.927908		
canopy	0.147876	0.450132	0.29900	1.000
moisture	-0.169369	0.143342	-0.01301	0.185
N	-0.144389	0.268068	0.06184	0.103
P	-0.058967	0.053720	-0.00262	0.028
pH	-0.153830	0.240305	0.04324	0.289
ERH	-0.500671	0.189227	-0.15572	0.173
root_col	-0.773694	-0.097025	-0.43536	0.166

	Coefficients[d]			
	Estimate	SE	t value	Pr(>|z|)
(Intercept)	1.53649	0.19630	7.937	3.03E-09
canopy	0.32746	0.06715	4.876	2.48E-05
ERH	-0.27527	0.14740	-1.867	0.0705
root_col	-0.44094	0.16240	-2.715	0.0103

[a] Residual standard error: 0.5624 on 34 degrees of freedom

[b] Multiple R-squared: 0.4785, Adjusted R-squared: 0.4324

[c] F-statistic: 10.4 on 3 and 34 DF, p-value: 5.291e-05

[d] lm(formula = Mv_SLA ~ Can + ERH + Root_col + 1)

Table 9D: ERH

	Model-averaged coefficients (conditional averages)				
	Estimate	SE	AdjustedSE	z value	Pr(>|z|)
(Intercept)	-8.50E-11	1.07E-01	1.09E-01	0.000	1.000
canopy	1.11E-01	1.11E-01	1.13E-01	0.982	0.326
moisture	-1.59E-01	1.65E-01	1.66E-01	0.958	0.338
N	1.63E-01	1.12E-01	1.13E-01	1.436	0.151
P	5.90E-02	1.09E-01	1.11E-01	0.533	0.594
pH	1.48E-01	1.80E-01	1.82E-01	0.815	0.415

	Confidence Intervals		Other	
	2.50%	97.50%	Coefficient	Importance
(Intercept)	-0.212694	0.212594		
canopy	-0.125281	0.195641	0.03518	0.317
moisture	-0.299981	0.188102	-0.05594	0.351
N	-0.144164	0.294413	0.07512	0.111
P	-0.099461	0.125407	0.01297	0.013
pH	-0.192712	0.282761	0.04502	0.045

	Coefficients[d]			
	Estimate	SE	t value	Pr(>|z|)
(Intercept)	-9.02E-11	1.06E-01	0.00	1.000
N	1.69E-01	1.07E-01	1.58	0.118

[a] Residual standard error: 0.5723 on 37 degrees of freedom

[b] Multiple R-squared: 0.02853, Adjusted R-squared: 0.0171

[c] F-statistic: 2.497 on 1 and 85 DF, p-value: 0.1178

[d] lm(formula = ERH ~ N + 1)

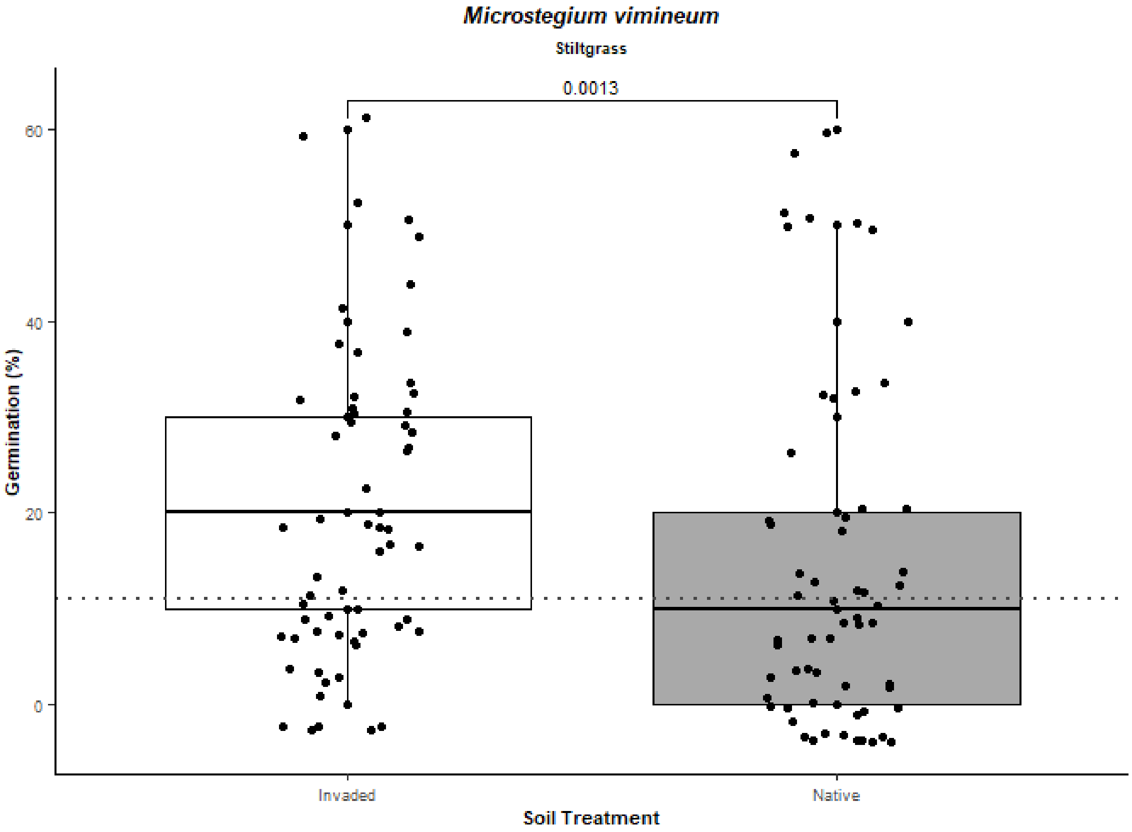
Table 3E. Root Colonization[a]

	Model-averaged coefficients (conditional averages)

APPENDIX II

SUPPLEMENTAL DATA FROM CHAPTER III

Appendix II Figure 17: *Microstegium* germination across soil treatments. P-value obtained from secondary pairwise t-test analysis, with replicates treated independently. Data points have been jittered for clarity. Dotted line indicates control group mean.



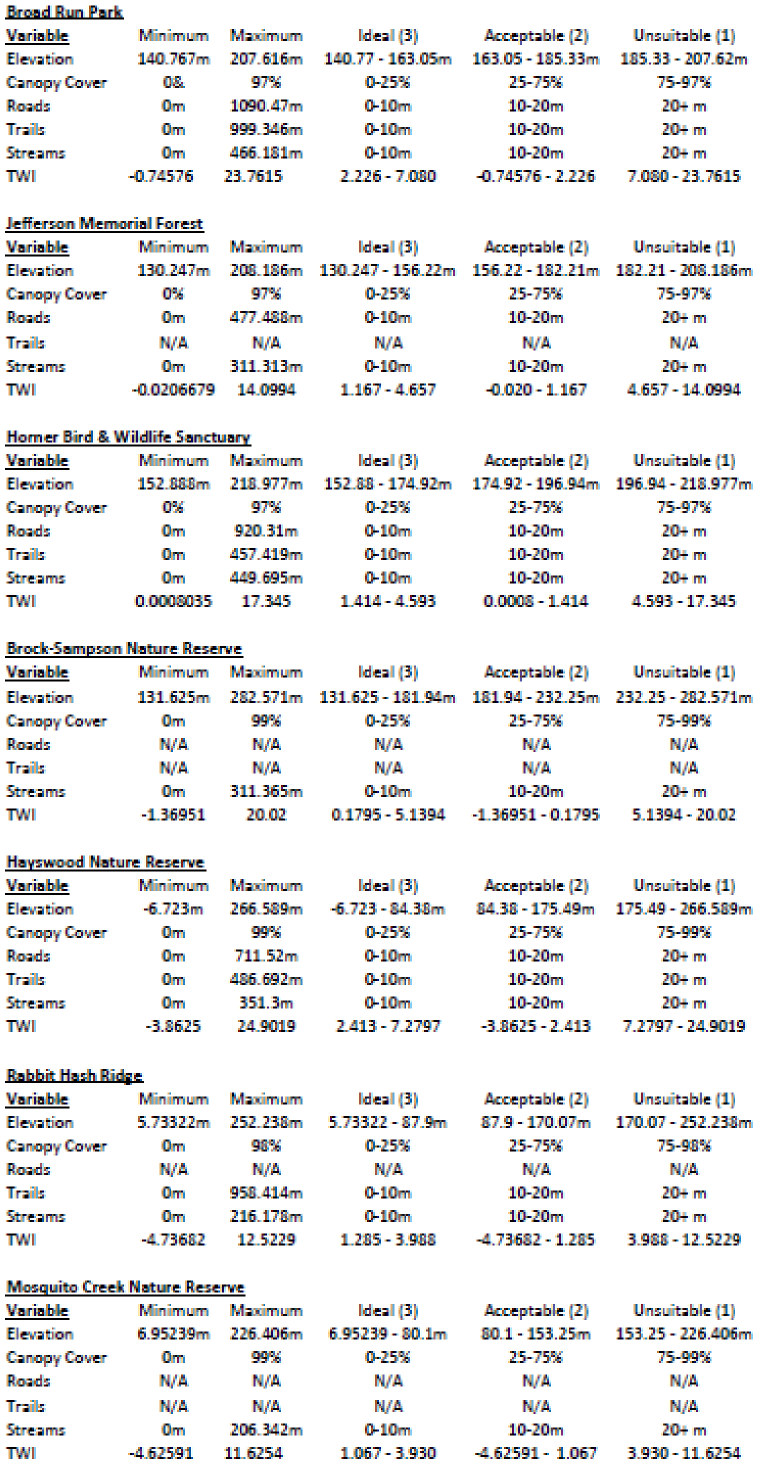
Appendix II Table 10: Mean germination, mortality, and growth responses for all native woody species.

Germination & Mortality		*Acer saccaharum*	*Fraxinus pennsylvanica*	*Quercus alba*	*Asimina triloba*	*Carya ovata*	*Quercus rubra*	*Cercis canadensis*	*Diospyros virginiana*	*Quercus velutra*	*Liriodendron tulipifera*	**Total Mean**
Percent Germination												
	Total	76%	59%	59%	57%	50%	20%	13%	7%	0.40%	0.29%	
	Native Mean	77%	57%	20%	50%	43%	20%	7%	10%	N/A	6.67%	32%
	Invaded Mean	77%	60%	21%	67%	53%	17%	17%	3%	10%	N/A	36%
	Control Mean	70%	60%	40%	50%	60%	30%	20%	10%	N/A	N/A	43%
Days to Germination												
	Native Mean	9.61	10.30	16.00	46.00	24.73	18.06	20.50	32.00	N/A	24.5	22.41
	Invaded Mean	9.18	10.30	15.00	45.00	32.53	23.00	20.00	31.00	14.33	N/A	22.26
	Control Mean	9.71	10.67	22.30	45.00	28.00	26.00	17.00	31.00	N/A	N/A	23.71
Seedling Mortality	**Native Mean**	0.378	0.153	0.057	0.197	0.047	0	0	0	N/A	1.00	0.204
	Invaded Mean	0.247	0.153	0	0.350	0.197	0	0.250	0	1.00	N/A	0.244
	Control Mean	0.143	0.167	0	0	0.167	0	0	0	N/A	N/A	0.060
Tree Seedling Growth		*Acer saccaharum*	*Fraxinus pennsylvanica*	*Quercus alba*	*Asimina triloba*	*Carya ovata*	*Quercus rubra*	*Cercis canadensis*	*Diospyros virginiana*	*Quercus velutra*	*Liriodendron tulipifera*	
	Plants Harvested	37	34	40	32	31	14	N/A	N/A	N/A	N/A	
Belowground Mass (g)												
	Native Mean	0.31	0.37	0.69	0.84	3.88	5.74	N/A	N/A	N/A	N/A	1.97
	Invaded Mean	0.36	0.38	0.95	0.86	3.52	5.57	N/A	N/A	N/A	N/A	1.94
	Control Mean	0.20	0.26	0.56	0.71	4.19	4.00	N/A	N/A	N/A	N/A	1.65
Aboveground Mass (g)												
	Native Mean	0.41	0.31	1.31	0.35	0.91	3.22	N/A	N/A	N/A	N/A	1.09
	Invaded Mean	0.41	0.39	1.63	0.37	1.01	4.26	N/A	N/A	N/A	N/A	1.35
	Control Mean	0.32	0.32	0.90	0.36	0.82	4.62	N/A	N/A	N/A	N/A	1.22
Stem Height (cm)	**Native Mean**	8.89	10.70	18.03	9.75	22.10	19.83	N/A	N/A	N/A	N/A	14.88
	Invaded Mean	8.89	12.67	20.82	10.29	21.00	27.40	N/A	N/A	N/A	N/A	16.85
	Control Mean	8.10	12.82	15.68	12.13	20.60	29.00	N/A	N/A	N/A	N/A	16.39

APPENDIX III

SUPPLEMENTAL DATA FROM CHAPTER IV

Appendix III Table 11: Habitat variable reclassification.



Broad Run Park

Variable	Minimum	Maximum	Ideal (3)	Acceptable (2)	Unsuitable (1)
Elevation	140.767m	207.616m	140.77 - 163.05m	163.05 - 185.33m	185.33 - 207.62m
Canopy Cover	0&	97%	0-25%	25-75%	75-97%
Roads	0m	1090.47m	0-10m	10-20m	20+ m
Trails	0m	999.346m	0-10m	10-20m	20+ m
Streams	0m	466.181m	0-10m	10-20m	20+ m
TWI	-0.74576	23.7615	2.226 - 7.080	-0.74576 - 2.226	7.080 - 23.7615

Jefferson Memorial Forest

Variable	Minimum	Maximum	Ideal (3)	Acceptable (2)	Unsuitable (1)
Elevation	130.247m	208.186m	130.247 - 156.22m	156.22 - 182.21m	182.21 - 208.186m
Canopy Cover	0%	97%	0-25%	25-75%	75-97%
Roads	0m	477.488m	0-10m	10-20m	20+ m
Trails	N/A	N/A	N/A	N/A	N/A
Streams	0m	311.313m	0-10m	10-20m	20+ m
TWI	-0.0206679	14.0994	1.167 - 4.657	-0.020 - 1.167	4.657 - 14.0994

Horner Bird & Wildlife Sanctuary

Variable	Minimum	Maximum	Ideal (3)	Acceptable (2)	Unsuitable (1)
Elevation	152.888m	218.977m	152.88 - 174.92m	174.92 - 196.94m	196.94 - 218.977m
Canopy Cover	0%	97%	0-25%	25-75%	75-97%
Roads	0m	920.31m	0-10m	10-20m	20+ m
Trails	0m	457.419m	0-10m	10-20m	20+ m
Streams	0m	449.695m	0-10m	10-20m	20+ m
TWI	0.0008035	17.345	1.414 - 4.593	0.0008 - 1.414	4.593 - 17.345

Brock-Sampson Nature Reserve

Variable	Minimum	Maximum	Ideal (3)	Acceptable (2)	Unsuitable (1)
Elevation	131.625m	282.571m	131.625 - 181.94m	181.94 - 232.25m	232.25 - 282.571m
Canopy Cover	0m	99%	0-25%	25-75%	75-99%
Roads	N/A	N/A	N/A	N/A	N/A
Trails	N/A	N/A	N/A	N/A	N/A
Streams	0m	311.365m	0-10m	10-20m	20+ m
TWI	-1.36951	20.02	0.1795 - 5.1394	-1.36951 - 0.1795	5.1394 - 20.02

Hayswood Nature Reserve

Variable	Minimum	Maximum	Ideal (3)	Acceptable (2)	Unsuitable (1)
Elevation	-6.723m	266.589m	-6.723 - 84.38m	84.38 - 175.49m	175.49 - 266.589m
Canopy Cover	0m	99%	0-25%	25-75%	75-99%
Roads	0m	711.52m	0-10m	10-20m	20+ m
Trails	0m	486.692m	0-10m	10-20m	20+ m
Streams	0m	351.3m	0-10m	10-20m	20+ m
TWI	-3.8625	24.9019	2.413 - 7.2797	-3.8625 - 2.413	7.2797 - 24.9019

Rabbit Hash Ridge

Variable	Minimum	Maximum	Ideal (3)	Acceptable (2)	Unsuitable (1)
Elevation	5.73322m	252.238m	5.73322 - 87.9m	87.9 - 170.07m	170.07 - 252.238m
Canopy Cover	0m	98%	0-25%	25-75%	75-98%
Roads	N/A	N/A	N/A	N/A	N/A
Trails	0m	958.414m	0-10m	10-20m	20+ m
Streams	0m	216.178m	0-10m	10-20m	20+ m
TWI	-4.73682	12.5229	1.285 - 3.988	-4.73682 - 1.285	3.988 - 12.5229

Mosquito Creek Nature Reserve

Variable	Minimum	Maximum	Ideal (3)	Acceptable (2)	Unsuitable (1)
Elevation	6.95239m	226.406m	6.95239 - 80.1m	80.1 - 153.25m	153.25 - 226.406m
Canopy Cover	0m	99%	0-25%	25-75%	75-99%
Roads	N/A	N/A	N/A	N/A	N/A
Trails	N/A	N/A	N/A	N/A	N/A
Streams	0m	206.342m	0-10m	10-20m	20+ m
TWI	-4.62591	11.6254	1.067 - 3.930	-4.62591 - 1.067	3.930 - 11.6254

Appendix III Table 12: Statistical Model Averaging results.

Variable	Coefficient	Stan. Error	Importance	LCL	UCL	p-value
Canopy Openness	6.9397336	1.528321	1.0000000	3.937807	9.941660	0.00001
Elevation	-0.7460481	1.339610	0.4138088	-3.375461	1.883365	0.57814
Road	-6.8435075	1.624314	1.0000000	-10.034320	-3.652695	0.00003
Stream	-4.7622723	1.547961	1.0000000	-7.803083	-1.721460	0.00214
Trail	-1.3614175	1.697535	0.5516668	-4.692244	1.969409	0.42307
TWI	-1.0159512	1.437552	0.4961840	-3.836786	1.804884	0.48025

Variable	Coefficient	Stan. Error	Importance	LCL	UCL	p-value
Canopy Openness	6.9397336	1.528321	1.0000000	3.937807	9.941660	0.00001
Road	-6.8435075	1.624314	1.0000000	-10.034320	-3.652695	0.00003
Stream	-4.7622723	1.547961	1.0000000	-7.803083	-1.721460	0.00214
Trail	-1.3614175	1.697535	0.5516668	-4.692244	1.969409	0.42307
TWI	-1.0159512	1.437552	0.4961840	-3.836786	1.804884	0.48025
Elevation	-0.7460481	1.339610	0.4138088	-3.375461	1.883365	0.57814

CURRICULUM VITAE

Kimberly Anne Koenig, MSSW, MBiol, PhD Fellow

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139 Life Sciences Building, Louisville, KY 40292
Email: kimberly.koenig@louisville.edu

Served 11 years in the mental health field before pursuing a PhD in plant and soil community ecology. Recognized in my current position as a patient and compassionate mentor for a diverse set of undergraduate students in both the field and in the lab. Also recognized for independence, self-motivation, time management, and effective verbal and written communication. Known for making professional connections and coordination of learning opportunities to gain needed skills. Experience with field and lab research, statistical analysis, GIS mapping, writing grant proposals, and identification of native and invasive plant species, as well as technical writing of manuscripts and presentation of scientific data. As a result of my research, I have extensive knowledge of soil science, conservation ecology, environmental biology, population and community ecology, invasion ecology, plant ecology and systematics, prescribed fire, and limnology.

Education

University of Louisville	August 2018 - Present
Master of Science in Biology	December 2022
Doctor of Philosophy in Biology	Graduation: May 2023
University of Louisville	Spring 2018
Northern Kentucky University	Fall 2017
University of North Carolina at Wilmington	Fall 2016 – Sum. 2017
Coursework in undergraduate Biology	
University of Louisville	Fall 2007 – Spring 2010
Master of Science in Social Work, Graduated May 2010	
University of Kentucky	Fall 2002 – Fall 2005
Bachelor of Arts in Psychology, Graduated Cum Laude December 2005	

Publications

Koenig, K. & Emery, S. M. 2021. Abiotic factors, not mycorrhizal associations, predict size and abundance of the Invasive grass *Microstegium vimineum*. The Journal of the Torrey Botanical Society 148(4):294-307.

Koenig, K. & Day, C. A. 2023. *Microstegium vimineum* habitat suitability analysis in the Kentuckiana region using geographic information system (GIS) modeling. Southeastern Geographer 63(2):183-201.

Koenig, K, Lee, B., & Emery, S. M. 2023. *Microstegium vimineum* invasion impacts germination and growth of native species through soil conditioning. – *submitted for publication*

Applicable Skills:

- Field research in Eastern deciduous forests and marine coastal systems
    - GPS navigation and data collection
- Laboratory research (plant and soil)
- Coordination of projects and people
- Statistical analysis
- Plant identification
- GIS mapping and modeling
- Invasive species monitoring
- Writing grant proposals
- Writing technical scientific manuscripts
- Verbal and written communication
- Scientific presentations
- Knowledge of soil science, conservation, and environmental biology
- Knowledge of freshwater and marine systems

Research Experience:

University of Louisville, Louisville, KY August 2018 to Present

PhD Dissertation

- preparation and submission of university and state program research grant proposals
- experimental design for field and laboratory-based studies
- communication and coordination with local land managers
- examination of data using statistical methods and R software
- preparation of spatial grids using GIS mapping software
- GIS mapping and modeling of potential dispersal pathways of an invasive species
- GPS navigation of field sites and mapping of invasive species populations
- collection of abiotic data (slope, aspect, canopy cover, soil temp. & moisture, air temp. & humidity)
- collection of biotic data (plant growth & biomass, soil samples, vegetation percent cover)
- identification of plant species present within field research plots
- germination and growth rate trials of native and invasive grass and woody species
- extraction and analysis of mycorrhizal extraradical hyphae
- analysis of percent root mycorrhizal colonization present in soil samples

UNCW Center for Marine Sciences, Wilmington, NC Summer 2017

Undergraduate Research Assistant, Coastal Plant Ecology Laboratory

- collection of biomass, sediment, and water samples from nearshore seagrass beds
- species identification, analysis, separation, and preparation of biomass samples
- preparation of sediment, water, and chlorophyll samples for nutrient analysis
- maintain cleanliness of all materials and laboratory space

Field Experience:

Northern Kentucky University, Highland Heights, KY Fall 2017

Plant Ecology

- determine species composition of deciduous forest site
- collect demographic data of woody species for long-term research analysis
- measurement of leaf water potential analysis and photosynthetic rate analysis
- measurement of relative water content and soil moisture in collected samples

Limnology

- perform stream system quality analysis
- perform water quality testing and analysis via various measurement tools
- identification and quantification of algal species present within the water column

University of North Carolina Wilmington, Wilmington, NC Spring 2017

Plant Systematics & Marine Botany

- identification of vascular plant species and marine algal species using local keys
- distinguish phenotypical features and terminology used in classification
- incorporation of sampling methods for sedentary organisms
- systematic comparison between local forest systems and wetland communities

Work Experience:

Bellarmine University, Louisville, KY Aug. 2021 – Dec. 2022

Adjunct Faculty, Part-time

Ecology Lab

- lead lectures, discussions, field research, and laboratory activities
- interact with students, answer questions, and assist with learning objectives

University of Louisville, Biology Department, Louisville, KY

Instructor of Record August 2021 – Present

Environmental Biology

- design course, syllabus, lectures, and interactive laboratory assignments
- preparation of assessment materials, quizzes, and examinations, and provide feedback on performance
- lead lectures, discussions, and hands on activities to fulfill learning objectives

Volunteer Graduate Teaching Assistant Aug. – Dec. 2019-2021

Plant Taxonomy

- guest lecture: Fagaceae, Sapindaceae, Juglandaceae, & Ulmaceae
- assist lab groups with learning plant identification and terminology
- collection of plant specimens and preparation for laboratory assignments
- mentor students, answer questions, and assist with learning objectives

Graduate Teaching Assistant Jan. 2022 – Present

Human Anatomy & Physiology

- lead lectures, discussions, and interactive laboratory assignments
- mentor students, answer questions, and assist with learning objectives
- provide evaluation and feedback on performance

Graduate Teaching Assistant Aug. 2018 – Aug. 2021

Principles of Biology Laboratory

- preparation of teaching materials, quizzes, and examinations
- lead lectures, discussions, and interactive laboratory assignments
- mentor students, answer questions, and assist with learning objectives
- provide evaluation and feedback on performance

Guest Lecturer
Biology: Current Issues and Applications
February 2019

- design and implement lecture focusing on principles within community ecology and climate change
- design and lead interactive learning activity & preparation of examination questions
- mentor students, answer questions, and assist with learning objectives

Previous Employment Experience:

Mental Health Therapy Services, Wilmington, NC — Oct. 2012 – Nov. 2016
Social Services & Case Management, Paris & Georgetown, KY — Oct. 2010 – Sep. 2012
Equine Industry & Retail Management, multiple locations — Aug. 2000 – Oct. 2010

Professional Development:

Forestry & Invasive Species Workshop, Purdue Extension — October 2018
Ecological Society of America Annual Meeting — August 2019
National Wildfire Coordinating Group RxCM Certification — September 2019

- NWCG S-130 Firefighter Training & NWCG S-190 Introduction to Wildland Fire Behavior
- NWCG L-180 Human Factors in the Wildland Fire Service
- FEMA IS-00100.c Introduction to Incident Command System
- FEMA IS 00700.b An Intro. to the National Incident Management System

Oak Woodlands & Forests Fire Consortium

- Fire & Fuels Monitoring Training — October 2019

GIS Technologies in Public Gardens — February 2021

Community Outreach:

Picture a Scientist, Crosby Middle School — February 2021
Harrison County Native Habitat Alliance — Aug. 2019 – Present

- Participate in monthly board meetings to develop, plan, and coordinate community-level events

The Nature Conservancy Volunteer Day — May 2019; Nov. 2019

- Invasive species removal, ecological conservation of glade system

Central Kentucky Riding for Hope, Lexington, KY — Aug. 2008 – Apr. 2009

- Grant preparation and program development

Awards Received:

- University of Louisville Biology Department
  - Beechmont Garden Award, $1000 — April 2020

Grants Received:

- Kentucky Academy of Science
  - Marcia Athey and Botany Fund, $2558.21 — March 2019
- University of Louisville
  - Graduate Student Council Research Fund, $489.53 — March 2019
  - Arts & Science's Undergraduate Mentoring Grant, $1000 — April 2019
  - Graduate Network in Arts & Sciences Research Fund, $250 — March 2020
  - Arts & Science's Undergraduate Mentoring Grant, $1000 — October 2020

Professional Presentations:

- Ecological Society of America August 2021
    - Abiotic factors, not mycorrhizal associations, are strong predictors of growth and abundance of the invasive grass *Microstegium vimineum.*
- Kentucky Academy of Science Annual Conference November 2020
    - Abiotic factors, not mycorrhizal associations, are strong predictors of growth and abundance of the invasive grass *Microstegium vimineum.*
- UofL Graduate Student Regional Research Conference February 2020
    - The role of mycorrhizal associations in promoting *Microstegium vimineum* invasion.

References: Contact information available upon request.